AN INVESTIGATION OF ASEPTIC AND ANTISEPTIC TECHNIQUES AS PRACTISED IN THE OPERATING THEATRES OF THE UNIVERSITY OF ALBERTA HOSPITAL

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The undersigned hereby certify that they have read and recommend to the School of Graduate Studies for acceptance, a thesis entitled "An Investigation of Aseptic and Antiseptic Techniques as Practised in the Operating Theatres of the University of Alberta Hospital,"
submitted by Maxwell William Nimeck, B.Sc.,
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ABSTRACT

Aseptic and antiseptic techniques as practised in the operating theatres of the University of Alberta Hospital were investigated under normal working conditions in the absence of any indication of an outbreak of infection.

General environmental factors were investigated by air sampling and by attempts to determine the effects of traffic, air conditioning, temperature and relative humidity on aerial contamination. Average counts varied from 1 to 20 micro-organisms per cubic foot of air, depending on sampling conditions. The effect of traffic was inconclusive, but there was a suggestion that traffic was influential in increasing aerial contamination. Air conditioning units caused a gradual increase in aerial contamination, probably because particulate matter was prevented from "settling out" when the air was recirculated. The maximum degree of aerial contamination occurred at indoor temperatures of between 65 and 75 degrees Fahrenheit and outdoor temperatures of approximately 50 degrees Fahrenheit. No relationship could be demonstrated between aerial contamination and relative humidity.

Specific environmental factors influenced by the duration of the operative procedure which were studied included masking of personnel, splash basins, operating table mats and sheets. Masks were always heavily contaminated on the inside, but respiratory organisms were shown to pass through the masks on only 3 occasions. Masks were not always sterile before use. Water from splash basins usually remained sterile or the number of organisms present was too few to count. One-fifth of the basins were contaminated and one-fourth of these showed a high degree of contamination. Fewer organisms were recovered from operating table mats after operations than before. Sheets were more heavily contaminated after the operative procedure than before.

Factors affecting cleaning which were studied included scrub water

for floors, mops and floors. Both mops and scrub water were always heavily contaminated. The degree of contamination decreased progressively throughout the day. Floors also showed progressive decrease in contamination. A much greater degree of contamination was demonstrated immediately after mopping than before.

Factors concerning anaesthesia which were studied included anaesthesia masks and endotracheal tubes. Anaesthesia masks were frequently heavily contaminated. Pathogenic micro-organisms were sometimes present. A decontamination procedure was effective if properly followed. Endotracheal tubes were usually contaminated. Pathogenic micro-organisms were frequently present. A modification of the decontamination procedure reduced the degree of contamination by one-half.

Incidental factors studied were stretcher blankets, various depots which might harbour micro-organisms, surgeons' hands, patients' skin and normal respiratory tract flora of personnel. Stretcher blankets were always contaminated. M. pyogenes var. aureus was contained in two-thirds of the cultures. Depots were usually contaminated depending on their location. Floors, walls and the base of the operating tables had the greatest degree of contamination. Control depots (sampled in the laboratory) showed only a slightly greater degree of contamination. Preoperative scrubs resulted in a 95% reduction in the flora of surgeons' hands. Cultures taken after the operative procedure showed a further slight decrease in flora. Preliminary cultures showed that preoperative skin preparation resulted in a 45% reduction in the patients' skin flora. The number of organisms was decreased further after the operative procedure when an iodine-alcohol treatment was used, but increased when Zephiran chloride was used for the preoperative skin preparation. The total nasopharyngeal carrier rate for M. pyogenes var. aureus was 44% for operating theatre personnel. Streptococcus pyogenes was recovered from the throat of only one individual.

The results of the investigation were discussed in relation to existing techniques and measures which might be adopted to reduce the degree of contamination.

THE UNIVERSITY OF ALBERTA

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INTRODUCTION

INTRODUCTION

The importance of cleanliness during operative procedures was appreciated long before the causative agents of infection were known. Ancient writings in Sanscrit discuss the importance of cleanliness of the operator's hands and the advantage of shortly clipped hair and beards. Before the development of surgical asepsis erysipelas, pyemia, septicemia and hospital gangrene were prevalent. Surgery was limited by the high mortality rate of 50%. Minor wound sepsis indicated by the presence of "laudable pus" was regarded as a sign of healing (Trent 1946). Burton and Homes investigated the transmission and contagiousness of purulent and surgical fever. White noted the similarity of the two conditions and advocated cleanliness as a means of controlling their spread. Semmelweiss was the first to prove that the transmission of infection could be prevented by a simple hand washing technique. Pasteur's discoveries of the universal distribution of micro-organisms and his germ theory of disease established a rational basis for further measures for the prevention of infection. Koch later isolated Streptococcus pyogenes which was the most serious cause of wound infection at this time. In 1881 Ogston isolated Staphylococcus aureus (Micrococcus pyogenes var. aureus) from suppurative lesions. In 1880 Lister developed his antiseptic surgical technique which included phenol dressings for the skin and instruments as well as a phenol spray. Von Bergman developed aseptic surgical techniques which gradually replaced antisepsis (Williams 1956), although the relative merits of both are still under discussion (Leading Article, Lancet, 1956). Bacteriologists by this time had shown that air was not the major source of infection and that many of the agents used in antisepsis were ineffective. Early

aseptic techniques included the following measures:

separation of clean and dirty cases,

sterilization of air by heat and filtration,

soap and water scrubs along with mercuric chloride or iodide,

soaks for the operating team,

sterilization of drapes by boiling,

closure of wounds without drainage,

(Trent 1946).

During the early twentieth century further developments such as the use of rubber gloves and masking showed the triumph of asepsis. During World War I the increased incidence of wound infection caused a return to chemical disinfection of wounds. The Second World War again saw increased wound infection, but this time the value of asepsis was recognized (McKissock et al., 1941; M.R.C. War Memorandum, 1944).

Prior to this period newer techniques such as serological typing made possible renewed studies concerning the transmission of infection. Wells and Wells (1936) established the importance of air as a means of transmission of infection. The development of chemotherapy and antibiotics reduced the importance of streptococcal infections which can be largely controlled by these agents, although streptococcal infections associated with drug-resistant staphylococci may be difficult to treat (Rountree, 1955). In their place, staphylococcal infections have become increasingly prevalent and this situation has been aggravated by the development of antibiotic-resistant strains of this organism.

Among the various bacteriological methods used in investigating staphylococcal infections, antibiotic sensitivity was the first used. Tube dilution (Lepper et al., 1953-1955) and sensitivity discs (Finland

and Haight, 1953), as well as the plate screening method (Rantz and Rantz, 1956) have been used. Serological typing has also been used (Brodie et al., 1956). Phage typing has been of special use (Williams and Miles, 1949; Williams and Rippon, 1952-1953; Rountree, 1953) since phage susceptibility is a strain-specific characteristic. Most workers used a combination of antibiotic sensitivity and phage in typing strains.

The importance of nasal carriers of Staphylococcus aureus and their relation to wound infection has been of interest (Miles et al., 1944; Williams and Miles, 1949). Spink (1954) and Landy et al. (1954) discussed the importance of carriers as a reservoir for infection. The increasing antibiotic resistance of strains of staphylococci has been demonstrated (Knight and Collins, 1955; Lowbury, 1955). Nasal carriers have also been cited as a possible source of infection in maternity hospitals (Knott et al., 1944). Colbeck (1949) has found the nasopharynx of infants to be a reservoir for staphylococci. Barber and Burston (1955) investigated the relation between nasal carriers among personnel and conjunctival carriers among infants. Other workers who investigated the carrier rate among personnel and infants in relation to infection rates include McGuiness and Musgrove (1949), Denton et al. (1950), Edmunds et al. (1955) and Sherman et al. (1956). Many workers including Lowbury (1955), Tulloch (1954), Gould and Allan (1954) and Barber and Burston (1955) recommended the treatment of carriers as a means of preventing the spread of hospital infections.

The increase in antibiotic resistance of Micrococcus pyogenes var. aureus, especially in the hospital environment, has been the cause of alarm to many hospital administrators. Duff and Murray (1953) warned of increasing clinical and bacteriological problems caused by widespread

antibiotic use. Among those suggesting control of the use of antibiotics were Clough (1955), The Lancet (Leading Article, 1956) and the Southern Medical Journal (Editorial, 1953). Newer antibiotics have been introduced including erythromycin (Forfar et al., 1955) and novobiocin (Rutenberg et al., 1956), but micro-organisms develop resistance to these agents. The only rational approach to the problem appears to be a restriction of the use of the newer agents for emergencies (Leading Article, Lancet, 1956; Leading Article, British Medical Journal, 1956).

Despite this confusion of literature concerning increased infection and carrier rates, several basic factors have become apparent. Howe (1954) stated that the development of infection depends upon the resistance of the host, virulence of the infecting organism and the size of the inoculum. The danger of infection is no greater than before, the present differences being the type and preponderance of the organisms involved. McDermott (1956) claimed that the incidence of healthy carriers which has been so widely reported has little to do with disease. He claimed that staphylococci have not increased their pathogenicity since the relative number of admittances of staphylococcal infections to hospitals has remained the same as in former years. There is little evidence of increased severity of staphylococcal disease or the spread of unusually virulent organisms. Instead the delicate balance in host - parasite relationship has been shifted because of a greater prevalence and spread of the infecting organisms. Modern therapy may be responsible for decreased host resistance because of the survival of patients less able to combat infection, drug therapy which lowers resistance and the frequency of skin punctures in therapy. The importance of decreased host resistance in staphylococcal infections has also been stressed by Elek (1956).

The congregation of susceptible hosts in hospitals is also of importance in the spread of infections. Because of the ubiquitous character of the organisms involved, McDermott stated that their exclusion from the hospital environment is impossible. The rational approaches to this problem are increasing continuous asepsis in all O.R. or ward procedures and investigation concerning means of increasing the host's defenses against infective organisms.

The possible sources of contamination which have been considered in operative wound infection are many and varied. Brewer (1915) discussed the importance of adequate sterilization of supplies and the use of antiseptics in preventing contamination from O.R.* supplies. Meleney (1935, 1940) stressed the importance of direct and indirect contact via the operators' hands and respiratory tract, the skin of the patient, instruments and materials used during operating, and the air as possible sources for streptococcal infections. Gardner (1937) stressed the importance of the air as a vector for infecting organisms suggesting the use of dust traps, masking, instrument canopies and the elimination of air currents to avoid postoperative infection. Hare and Willets (1941) recognized the importance of dust control in preventing the spread of staphylococcal and streptococcal infections. Urkov (1945) suggested that operating techniques are adequate but that asepsis should be practised in wound dressing. Blair (1948) listed the respiratory tract and hands of the operator, the skin of the patient, instruments and the air as sources of contamination. Walter (1952) said that the construction of operating rooms is of great importance in proper aseptic techniques. Clark et al. (1952) discussed the importance of aerial contamination from carriers, air and dust from bedclothes.

* Operating Room

Colebrook (1955) suggested the oiling of floors and bedclothes along with positive pressure ventilation as aids to general aseptic measures. Shooter et al. (1956) also suggested the use of air conditioning in operating rooms for reducing aerial contamination. Hare (1956) stressed the importance of certain respiratory carriers in the spread of infections. Colbeck (1956) reviewed the importance of fomites especially in relation to bedding and hand washing facilities. The multiplicity of possible sources of contamination has been recognized by such workers as Dolman (1956), and Starkey (1956). The latter stated that there is no well-defined source or reason for hospital staphylococcal infections and every hospital should regard itself as being equally susceptible to infections. Because of the ubiquitous character of the organisms involved all means of preventing infections should be employed and a systematic rechecking of all existing measures should be carried out.

EXPERIMENTAL

GENERAL ENVIRONMENTAL FACTORS

(a) "Air Sampling"

Historical Introduction

Pasteur was the first to show that micro-organisms were always present in the air. Lister, influenced by Pasteur's ideas, devised a continuous phenol spray to prevent aerial contamination of surgical wounds. Chaplin (1914) claimed that expired air was normally sterile. Pflugge studied droplet infection showing that expired droplets over 0.1 mm. in diameter fell quickly and were thus unimportant in the transfer of respiratory infections. Wells and Wells (1936) showed that Pflugge's work was incomplete in that droplets less than 0.1 mm. in diameter remained suspended for hours and a large proportion of these evaporated, leaving a residue of organic material containing bacteria (droplet nuclei) which could remain suspended 2 or more days before settling. Droplets, droplet nuclei and dust were shown to be the important factors contributing to aerial contamination. The question of what part certain individual factors play in contamination is still unsettled. Hart (1946) claimed that aerial contamination has become important again because of adequate control of other direct and indirect sources of contamination. Longer more traumatizing operations on debilitated patients as a result of surgical advances may also be a contributing factor. Hart (1938) included inadequate masking, clinical carriers, personnel with respiratory infections, the number of occupants in operating rooms and the type of activity as possible factors contributing to aerial transmission of pathogenic micro-organisms.

[illegible]

[illegible]

[illegible]

[illegible] [illegible]

Bourdillon and Colebrook (1946) stated that blankets and dressings may be important sources of air-borne pathogens. Hamburger and Green (1946) stressed the importance of the carriers in the transmission of streptococcal infections. Nose blowing, hands and handkerchiefs were listed as possible vectors for the spread of streptococci. Pulvertaft (1947) stressed the importance of dust in air-borne infections. He recommended ventilation and oiling for dust control. Colebrook and Cawston (1948) showed that outside air contains bacteria but relatively few pathogens compared to the air of an occupied room. They recommended ventilation with outside air as a means of reducing the numbers of air-borne bacteria. Bourdillon et al. (1948) listed the following sources of air-borne bacteria: hair of personnel, respiratory tracts of personnel, skin of patients, operators' hands, textiles used in operating rooms, air sucked in from below operating room doors and windows.

Girdleston and Bourdillon (1951) have set the following arbitrary limits of aerial contamination:

Minor operations and dressings ... 20 organisms per cu. ft.

Major operations on tissues of normal resistance to infection .... 10 organisms per cu. ft.

Burns and operations on tissues of low resistance to infection .... 0.1 to 2 organisms per cu. ft.

These figures, representing total numbers of bacteria and not necessarily pathogens, were arrived at by statistical evaluation of aerial contamination and infection rates. The ideal situation would, of course, be an atmosphere entirely free of bacteria.

Various methods have been used to control air-borne bacteria, including chemical disinfection, heat treatment, ultraviolet radiation, air conditioning, air traps and dust control by oiling.

Chemical disinfection was used long before the spread of infection by aerial routes was established, e.g. burning sulphur was used during the Middle Ages. The first rational use of chemicals was Lister's phenol spray which was used to combat air-borne contamination in operating rooms. Phenol has long since been replaced by less irritating chemicals. Formalin produced by the action of potassium permanganate on paraformaldehyde is still used for disinfection of air in unoccupied areas. Hypochlorites have been used as a 1% sodium hypochlorite spray or by boiling an aqueous solution with sodium acid phosphate buffer. Bleaching powder has been reacted with water and carbon dioxide (Bourdillon et al., 1948). Because hypochlorites are corrosive their use is limited to confined areas with poor ventilation. Propylene glycol has been dispersed by vaporization using a hot plate. This agent is effective but condenses on walls and windows (Robertson, 1946). Triethylene glycol has been used by vaporization but its odor is a limiting factor. Organic acids such as lactic or leavulenic acid have been used by spraying but they tend to be irritating. Al iphatic alpha-hydroxy carboxylic acids such as alpha-hydroxy methyl butyric acid have also been used. MacKay (1952) described the use of hexylresorcinol as heat-generated vapour. The drug is ineffective for spores. Nash (1951) stated that relative humidity and solubility limits affect the efficiency of chemicals since saturation rather than concentration is important. If the humidity is too high the chemical is diluted. If the humidity is too low the substance may precipitate. Robertson (1946) described a glycostat for determining and regulating the concentration of disinfectant vapours. Ingraham (1950) listed toxicity, offensiveness, methods of dispersal (sprays, heat, vaporization

at or near room temperature) and difficulties in maintaining vapour concentration as factors which limit the usefulness of aerial disinfectants. The conflicting results shown in field trials are due to the difficulty in establishing controls.

Heat has been used in aerial decontamination as dry heat or steam. Dry heat is usually applied by means of electric furnaces internally to the area to be treated or Bunsen burners may be used as a source of heat in emergencies. The temperature required depends on the type of bacteria, size of particles, presence of protein or other organic material and sporing. Temperatures of 175°C. to 300°C. have been used (Bourdillon et al., 1948). Steam has been circulated through rooms. The number of bacteria is reduced appreciably but sterilization is impossible by this means. Both types of heat treatment are limited to areas which can be sealed off during treatment. Corrosion is also a limiting factor especially with steam. After treatment progressive recontamination of the air readily occurs.

Of the many means of controlling air-borne micro-organisms ultraviolet radiation has received the most attention. Gates (1930) determined the effect of ultraviolet radiation on Staphylococcus aureus. Major and Wilder (1937) determined the effects of wavelength, exposure time, velocity, dust and relative humidity on the efficiency of ultraviolet lamps. Hart (1937-1938) claimed that the air of operating theatres is highly contaminated with pathogenic bacteria and that this air is an important source of contamination for every operative procedure. He recommended the use of low vapour pressure mercury lamps which emit rays of 2537 A° for the control of "unexplained infections" (1941-1942). Hart used exposed Petri plates, infection rates and postoperative temperature elevation as criteria in his evaluation. Wells and Wells (1936) used their air centrifuge in

determining the action of ultraviolet radiation on bacteria contained in droplet nuclei. Robertson et al. (1939-1940) evaluated the use of ultraviolet lamps in air ducts and as a radiant curtain compared to ventilation. Koller (1939) discussed the use of ultraviolet lamps in air ducts and the effect of relative humidity on their efficiency. Kraissl et al. (1940) used exposed Petri plates in a test chamber to determine the efficiency of ultraviolet lamps for the disinfection of operating room air. Rentschler et al. (1941, 1942) in their studies on aerial and deposited _E. coli_, showed that ultraviolet radiation is most effective for actively growing organisms. Cruickshank (1947) discussed ultraviolet radiation as a means of controlling infection spread by droplet nuclei. Anderson (1947) discussed the use of ultraviolet radiation in the elimination of air-borne infection; the prevention of cross-infection and as an adjunct to aseptic techniques. Frazer (1946, 1944) discussed the relative advantages of overhead ultraviolet lamps, lamps focused on the upper parts of walls, in air ducts and local or spot lamps. He stated that routine use of ultraviolet lamps during operations does not contribute to visceral irritation. Bourdillon and Lidwell (1948) stated that ultraviolet radiation is more effective for bacteria distributed by moisture droplets than for those contained in dust particles. Thomas et al. (1948) recommended the use of dust filters to reduce dust-borne organisms when ultraviolet lamps are used to reduce aerial contamination. Barrier radiation (batteries of lamps surrounding doorways of cubicles) requires high intensity units and little traffic to prevent air currents. Overhead or spot lamps must not cause irritation of the operators' or patients' skin. Air duct units require controlled air

currents. Because of these disadvantages the use of radiation is largely confined to units in air ducts in combination with dust filters and forced air conditioning. Goodman et al. (1949) stated that air conditioning alone increases aerial contamination, but is beneficial when combined with ultraviolet radiation and dust control.

Relative humidity and dust are important factors in the control of air-borne micro-organisms. Lowbury (1954) recommended a relative humidity of 50-60% when combined with ultraviolet radiation. Dunklin and Puck (1948) stated that the mortality rate of bacteria in air depends on relative humidity, the death rate being highest at 50%. The lethal effect of moisture is said to be due to impurities such as sodium chloride in the air-borne droplets. Taylor (1955) stated that a relative humidity of 50% to 55% is most effective in the presence of sodium chloride. Lidwell and Lowbury (1950) showed that relative humidity also has an effect on survival rates of dust-borne bacteria. Loosli (1948) stated that floors and bedclothes are the important factors in the spread of dust-borne micro-organisms. He recommended oiling as a control measure. Anderson et al. (1944) recommended oiling of floors (1 gallon of spindle oil per 1000 square feet of floor surface) for controlling respiratory infections. Clayton and Robertson (1945) also recommended oiling of floors. They described the use of an emulsion containing mineral oil and Fixanol C (cetyl pyridinium bromide) for oiling of bedclothes. Duguid and Wallace (1948) stated that dust from clothing may be associated with the spread of infections.

<u>Operating Theatres</u>

All operating theatres in the University of Alberta Hospital are

situated on the fifth floor of the north wing. Operating theatres are connected by a corridor which is separated from the main corridor by swinging doors. Traffic control regulations are posted but these regulations are not rigidly enforced. Use of the ambulance elevator is not restricted and it may be used for general traffic when other elevators are out of order, resulting in greatly increased corridor traffic at such times.

All doors of operating rooms open directly onto the corridor and are frequently left open throughout the operating day.

Windows are equipped with screens and draught guards. They are usually left open in warm weather except in operating theatres I, II and III, which were equipped with air conditioning units late in the course of this investigation.

There is no apparent control of traffic in operating theatres. The circulating nurse may enter an operating room several times to obtain supplies. Since there are no observation rooms, medical students must enter the operating rooms for teaching purposes. All persons entering operating theatres must be properly capped and masked. Only operators, assistants, instrument nurses and sponge nurses wear sterile gowns. Only operators and assistants wear overboots which were provided during this investigation, although this practice is not enforced. Nurses formerly wore ward uniforms, but are now provided with special uniforms. Persons may occasionally be seen entering operating theatres with street clothes and shoes.

Methods and Materials

Air Sampling Devices

Several types of sampling devices have been used in determining the spread and control of air-borne micro-organisms. They include Wells' and Wells' centrifuge tube (1936), various washing or bubbling devices, atomizers, and impinging devices. Among the latter type there are the funnel aeroscope, radial jet samplers, various types of slit samplers (Bourdillon et al., 1948; Dickes and Wilson, 1954) and the duplex electrostatic sampler (Luckiesh, Taylor and Halliday, 1946, 1947, 1949). The General Electric Bacterial Air-sampler was used for all tests in this series (Appendix A). This apparatus has a capacity of 0.5 cubic feet of air per minute.

Samples consisted of a pair of cultures on blood agar plates representing negative and positive charged particles respectively. The cultures are called "sampler plates" in this report. Cultures were also obtained by exposure of blood agar plates to collect bacteria settling out of the air. These plates are called "deposit plates". All cultures were incubated aerobically at 37.5°C. for 48 hours before counting colonies by the use of a Quebec Colony Counter.

Preliminary Experiments

Preliminary experiments to determine optimum sampling volume for the Electrostatic Air Sampler were conducted in room M 86 of the University of Alberta. The room is a large student laboratory, with the following dimensions: 57.5 ft x 45.5 ft x 13.5 ft. The total

volume of the room is 35,319 cu. ft. Samples were taken of $2\frac{1}{2}$, $7\frac{1}{2}$ and 30 cu. ft volumes of air. Samples of air were taken when students entered, in the presence of students, and after students had left the laboratory. One sample was also taken in M 71, a smaller student laboratory with the following dimensions: 27 ft x 15 ft x 13.5 ft. The total volume of this laboratory is 5,467 cu. ft. Incubation of sample plates was as previously described.

The results are shown in Table 1 A.(page 18). Organisms which grew on the sample plates included aerobic spore bearers, diphtheroids, Streptococcus viridans, Neisseria, moulds, Actinomycetes, Gaffkya, Sarcina and Micrococci. Hemolytic and indifferent Micrococcus pyogenes var. aureus and albus were included in the latter group. The total colony counts were in the neighborhood of 300 colonies for samples of 25 or 30 cu. ft of air. It was, therefore, decided to use 30 cu. ft volumes, requiring a one-hour sampling time, as a standard for sampling air in the hospital.

The results show a low order of contamination in the unoccupied laboratory with a significant rise in flora after students entered. The higher counts shown in samples taken with students present for half of the sampling period were probably due to the increase in activity during their exit from the laboratory. The count of aerial bacteria was reduced within an hour after students had left, but did not return to the original level during the test period.

Two preliminary sets of air samples were taken in operating rooms labelled operating theatres III and II. Using the electrostatic air-sampler, the first sample, at 5:00 to 6:00 a.m., was controlled by an automatic timing device. Notes were taken concerning the type of

Determination of Optimum Sample Volumes for Electrostatic Bacterial Air-sampler

Date, Sampling Conditions and Initial Time	Number of Colonies per Sample					Average no. of organisms per fu. ft of air
	$2\frac{1}{2}$ cu. ft	$7\frac{1}{2}$ cu. ft	15 cu. ft	30 cu. ft	Total	
Sept. 31, 1955 M 71						
11:00 a.m. Lab. empty			29		29	1.93
M 86						
12:30 p.m. Lab. empty	8	10	13		37	1.48
2:30 p.m. Students 1 hr.	16	55	172		243	9.72
4:00 p.m. Empty $\frac{1}{2}$ hr.	17	22	27		66	2.64
Oct. 1, 1955 M 86						
10:55 a.m. Lab. empty 5 min.	11	58	33		102	2.08
Oct. 3, 1955 M 86						
8:45 a.m. Lab. empty	.4	11	32		47	1.04
9:30 a.m. Students $\frac{1}{2}$ hr.	25	57	103		185	7.40
1:00 p.m. Students present $\frac{1}{2}$ time				305	305	10.17
Oct. 5, 1955 M 86						
2:00 p.m. Students present $\frac{1}{2}$ time	11	114	90		215	8.60
Nov. 25, 1955 M 86						
3:00 p.m. Students $\frac{1}{2}$ hr.				359	359	11.97
4:00 p.m. Students present $\frac{1}{2}$ time				335	335	11.17
Averages:						
Lab. empty (before students)						1.48
Students						9.70
Students present $\frac{1}{2}$ of time						9.98
Lab. again empty (after students)						2.36

operation, time, number of persons present and the location of doors and windows. The counts of these samples, listed in Table 1 B, (page 20), show a significant rise in bacterial flora of the air during occupation.

The next series of air samples consisted of 30 cu. ft volumes of air taken in the unoccupied theatre (at 5:00 to 6:00 a.m.), during the first case (at approximately 9:00 to 10:00 a.m.) and after the case (at approximately 11:30 a.m. to 12:30 p.m.). The results of these cultures are given in Table 2 A (page 21). Deposit plates were also taken by exposing blood agar plates in the operating room overnight until 9:00 a.m., from 9:00 a.m. until approximately 11:30 a.m. and from 11:30 a.m. to 12:30 p.m. The results of these cultures, recorded as the number of particles settling out on a blood agar plate per hour, are given in Table 2 B (page 22). Missed results in the "sampler" plates were due to the sampler being turned off. Missed results in "deposit" plates were due to drying of the culture medium. Organisms which grew on the culture plates included aerobic spore bearers, diphtheroids, Neisseria, coliforms, Actinomycetes, moulds, Streptococcus viridans, Gaffkya, Sarcina and Micrococci, including Micrococcus pyogenes var. aureus.

Because there was a greater degree of aerial contamination (22.4 organisms per cu. ft) in operating theatre III when the doors were open during the operative procedure, it was decided that a series of air samples should be taken from the north end of the corridor (near O.R. II) and from the centre of the corridor of the operating wing (near O.R. III). Samples of 30 cu. ft of air were taken at approximately the same times as those in the previous series. The results are shown in Table 2 C (page 23). The counts were higher for the samples

TABLE 1 B

Comparison of Degree of Bacterial Contamination of Operating Room Air Before and During Occupation

Date 1955	Operating Theatre	Time of Case	Time of Sampling	No. of organisms per cu.ft of air	
				Theatre Unoccupied	Theatre Occupied
Oct. 28	III	8:00-11:30 a.m.	5:00-6:00 a.m.	1.43	
			9:30-10:30 a.m.		23.33
Oct. 31	II	8:15-11:30 a.m.	5:00-6:00 a.m.	0.20	
			11:30 a.m. - 12:30 p.m.		10.00
			Average	0.82	16.67

TABLE 2 A

Degree of Bacterial Contamination of Operating Room Air Determined by Electrostatic Bacterial Air-sampler

Date 1955	O.R. No.	Theatre Unoccupied (5 - 6 a.m.)	During First Case		After Case	
		No. organisms per cu.ft air	Time of Sampling	No. organisms per cu.ft air	Time of Sampling	No. organisms per cu. ft of air
Dec. 1	III	0.30	9:00-10:00	22.8	12:15-1:15	-----
Dec. 2	III	2.67	9:00-10:00	----	11:00-12:00	10.17
Dec. 6	VII	0.23	9:00-10:00	----	12:00-1:00	11.5
Dec. 7	VII	0.46	9:00-10:00	----	12:00-1:00	-----
Dec. 8	VII	0.60	9:00-10:00	----	11:00-12:00	26.43
Dec. 9	V	----	9:00-10:00	10.2	-----------	-----
Dec. 13	VI	2.63	9:15-10:15	----	11:15-12:15	11.27
Dec. 14	III	0.60	9:00-10:00	----	11:30-12:30	-----
Dec. 15	II	0.93	9:00-10:00	----	10:15-11:15	6.9
Dec. 16	III	0.63	9:00-10:00	18.4	11:30-12:30	16.2
	Average	1.01		17.1		13.73

TABLE 2 B

Bacteria Settling out of Air in Operating Rooms Determined by Deposit on Exposed Culture Plates

Date 1955	O.R. No.	Theatre Unoccupied		During First Case		After Case	
		Time of Sampling	No. organisms per hour	Time of Sampling	No. organisms per hour	Time of Sampling	No. organisms per hour
Dec. 2	III	3:00pm-9:00am	13.4	9:00-11:00	67.5	11:00-2:30pm	71.5
Dec. 6	III	5:30pm-9:00am	11.5	9:00-11:00	----	11:00-3:00pm	45.3
Dec. 7	VII	3:30pm-9:00am	----	9:00-10:30	54.6	10:30-2:00pm	56.6
Dec. 8	VII	2:00pm-9:00am	42.0	9:00-10:00	67.0	10:00-2:00pm	68.0
Dec. 9	V	3:00pm-9:00am	4.8	9:00-10:00	52.0	------------	----
Dec.13	VI	6:30pm-9:15am	25.3	9:15-10:30	38.4	10:30-5:00pm	24.0
Dec.14	III	5:00pm-8:45am	3.1	8:45-11:00	39.6	11:00-3:00pm	28.5
Dec.15	II	3:00pm-8:45am	4.7	8:45-10:15	27.3	10:15-2:45pm	18.7
Dec.16	III	6:45pm-8:45am	8.3	8:45-11:00	72.0	11:00-2:00pm	51.6
		Average	14.1		52.3		45.5

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[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	----	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	----	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
----	----	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]		[illegible]		[illegible]	[illegible]		

TABLE 2 C

Degree of Bacterial Contamination of Air in Corridors of Operating Wing

Date	Location	Number of organisms per cu. ft of air		
		5:00-6:00 a.m.	9:00-10:00 a.m.	approximately 11:30 a.m. - 12:30 p.m.
Dec. 12/55	Centre of Corridor (near O.R. III)	2.93	33.0	30.3
Dec. 22/55		2.67	----	----
Dec. 23/55		1.3	20.3	14.9
Dec. 29/55		3.2	30.27	20.3
Jan. 4/56		3.0	48.0	31.7
	Average	2.6	32.9	24.3
Dec. 30/55	North end of corridor (near O.R. II)	0.93	14.53	12.73
Dec. 31/55		1.93	8.33	28.83
Jan. 6/56		1.40	19.67	19.33
Jan. 26/56		0.8	20.57	21.57
Jan. 27/56		1.3	18.30	10.77
	Average	1.27	16.28	18.64

taken from the centre of the corridor than for those taken from the end, probably because of differences in amount of traffic. Counts were almost double those obtained in operating rooms during the first case.

Controls

In order to have some basis for comparison for air samples from operating rooms, cultures of air were sampled in rooms M 86 and M 71 of the University of Alberta, the student laboratories used for the initial tests. Air samples were obtained hourly by use of the Electrostatic Bacterial Air-sampler and by exposure of blood agar plates. Culturing methods were the same as those previously employed. The results are given in Table 3 A (page 25). The number of organisms per cu. ft of air was much higher in the large laboratory with a large number of students than in the smaller laboratory with fewer students present.

A second set of hourly air samples was obtained of outdoor air by the same methods. Sampling apparatus was placed on window ledges, approximately 4 feet above ground level, on the east side of the east wing of the Medical Building. The results are shown in Table 3 B (page 26). Organisms recovered from this set of air samples were the same as those recovered in the laboratory, except that Micrococcus pyogenes var. aureus, Streptococcus viridans or Streptococcus pyogenes were present in the air in the laboratory and these organisms were not found in air taken outside the building.

TABLE 5 A

Degree of Bacterial Contamination of Air of Student Laboratories

Date	Location and Sampling Conditions	Initial Sampling Time	Method of Sampling: Electrostatic Air-sampler	Method of Sampling: Deposit
			No. of organisms per cu. ft of air	No. of organisms settling per hour
Nov. 6/56	M 86. Lab. unoccupied. Approx. 60 students entered at 1:25, remained until 4:35	12:27	2.30	15.0
		1:29	11.17	55.5
		2:52	14.37	87.0
		3:52	15.03	76.0
		4:52	5.67	45.0
	Average		9.71	55.7
Nov. 8/56	M 71. Lab. unoccupied. 17 students entered at 1:25, remained until 3:30	1:00	2.33	5.0
		1:30	2.20	27.0
		2:30	3.90	28.0
		3:30	2.83	21.0
		4:30	1.47	6.0
	Average		2.55	17.4

TABLE 3 B

Degree of Bacterial Contamination of Outside Air

Date	Location of Sampler	Sampling Time	No. of organisms per cu. ft of air	No. of organisms settling per hour
Nov. 23/56	Window ledge, centre east wing	9:23-10:30	7.49	486.5
		10:30-11:35	8.61	299.1
		11:35-12:35	12.67	440.0
		12:35-1:30	11.35	328.3
		1:30-2:32	8.77	274.8
		2:32-3:30	8.96	206.9
		3:30-5:45	13.24	158.7
		Average	10.16	313.5

Test Series

This series of air samples was obtained during 105 cases representing 35 operating days, using the General Electric Electrostatic Bacterial Air-sampler and by exposure of 90 mm. culture plates. The medium used for all cultures was blood agar containing 5% sheep's red blood cells. Cultures were incubated for 48 hours at 37.5°C. before counting. The first samples, using the Electrostatic Air-sampler, were taken at 5:00-6:00 a.m. in the unoccupied theatre using an automatic timing device. Samples were taken before the first case from approximately 7:30 a.m. until the first case was started. Air samples were then taken hourly during each case, between cases (during cleaning) and after the last case. Deposit samples obtained by exposure of culture plates were taken at the same times except for the initial plates which were exposed from the evening before when the timing device on the air sampler was set until between 7:30 and 7:45 of the test day. The air sampler was located in a corner of the operating room at the approximate level of the operating table. Culture plates for deposit samples were placed against the opposite wall from the air sampler, usually on the cupboard used for anaesthetic supplies, to avoid draught from the sampler. Complete chronological records were also kept of the type and length of operations, techniques, traffic, temperature, relative humidity, sampling times, agents used, possible breaks in technique, etc. The results of air sampling using the air sampler are given in Table 4 A (page 28), while Table 4 B (page 29) shows counts obtained by exposure of blood agar plates. The average counts for the sampler plates are presented graphically in Figure 4 C (page 30).

TABLE 4 A

Degree of Bacterial Contamination of Air in Operating Rooms

Determined with G.E. Electrostatic Bacterial Air-sampler

TABLE 4 A

Degree of Bacterial Contamination of Air in Operating Rooms Determined With G.E. Electrostatic Bacterial Air-sampler

Date 1956	O.R.	Numbers of organisms per cubic foot of air											
		Theatre Unoccupied 5 - 6 a.m.	Before First Case	During First Case	Before Second Case	During Second Case	Before Third Case	During Third Case	Before Fourth Case	During Fourth Case	Before Fifth Case	During Fifth Case	After Final Case
Jan. 5	II	0.7	25.5	13.7	19.2	5.9							14.2
12	II	0.5	15.6	15.2	28.3	25.8							15.8
28	VII	0.8	9.5	9.5	10.9	8.0							5.6
Feb. 2	VII	0.9	10.9	3.9	15.3	19.8	19.9	28.7					7.9
9	III	0.3	12.3	16.5	33.9	8.3							16.6
16	V	0.6	5.6	6.4	10.3	5.9	11.1	18.9	14.2	15.7			13.8
23	VII	0.8	6.9	13.3	28.4	15.9	33.4	21.5					15.7
Mar. 1	VI	0.7	9.5	4.9	10.4	15.2	11.3	8.2					4.8
8	V	0.4	27.1	20.1	29.8	12.1	31.5	22.3					21.3
15	III	0.3	27.1	25.1	27.2	16.9	19.5	8.5					16.0
22	I	1.8	12.5	15.5	22.4	12.8							8.9
29	III	1.9	6.2	4.1	6.8	9.0	11.2	4.6	16.2	4.1	7.5	14.2	8.2
May 17	V	0.6	17.5	13.4	26.0	13.2	15.7	8.0	9.8	15.6	22.0	15.2	13.2
24	VII	1.9	32.2	18.1	19.1	8.4	21.7	10.8	17.2	10.3			8.4
29	VI	1.3	29.0	9.0	17.5	7.2	13.9	12.3	19.3	15.7			21.7
30	II	1.0	15.4	15.7	22.1	15.2	12.1	11.4	20.0	20.0	6.6	10.6	7.9

(continued on next page)

TABLE 4 A

Degree of Bacterial Contamination of Air in Operating Rooms

Determined with G.E. Electrostatic Bacterial Air-sampler

Date 1956	O.R.	Numbers of organisms per cubic foot of air											
		Theatre Unoccupied 5 - 6 a.m.	Before First Case	During First Case	Before Second Case	During Second Case	Before Third Case	During Third Case	Before Fourth Case	During Fourth Case	Before Fifth Case	During Fifth Case	After Final Case
June 5	I	0.8	15.4	13.1	10.4	5.5	12.2	13.1	16.4	7.7			6.8
6	I	0.6	22.8	14.1	14.3	4.5	10.9	5.3					6.8
26*	II	0.7	27.2	17.5	28.8	16.9	19.6	11.6	16.7	11.4	25.9	9.3	19.4
27	II	0.4	19.6	11.3	11.1	8.9	21.4	6.9	18.0	16.8			27.9
July 4	VI	1.8	9.3	16.3	11.3	26.3							13.9
5*	II	1.6	52.2	12.5	23.4	8.0	32.8	9.8					3.3
25*	II	0.5	21.2	14.3	20.8	12.4							19.3
Aug. 8*	II	0.5	26.7	21.9	45.3	22.1	27.2	25.8					14.9
9*	II	0.7	21.4	5.6	18.8	9.8	26.3	15.6					18.9
16	VI	1.5	27.4	7.2	28.0	21.0							12.7
17	VI	1.9	26.6	14.2									16.8
22	V	0.3	37.1	19.5	29.5	13.9	26.8	11.2					22.9
23	V	0.5	25.3		22.2	17.9	23.4	16.8	12.7	7.3	11.2	25.7	35.5
Sept. 6	VII	0.4	24.2	30.7	29.0	31.4	34.8	17.6	39.9	36.1			17.5
7	VII	0.4	21.9	18.2	23.0	8.8	27.4	23.9	23.2	15.2	27.2	16.1	19.8
12*	III	0.7	21.9	9.8	17.9	8.1	23.9	12.1	23.2	13.2			10.8
13*	I	0.3	15.5	12.2	23.1	19.6	32.1	21.0					15.3
25*	I			12.5	16.2	19.5							13.6
26*	III	4.0	10.9	8.2	18.8	14.9	14.3	7.5					13.7
Averages		0.9	19.5	16.5	21.2	13.8	21.2	16.8	19.7	14.4	14.9	15.6	14.3

* Air conditioning unit in use

TABLE 4 B

Bacteria Settling out of the Air in Operating Rooms

Determined by Exposure of Culture Plates

TABLE 4B

Bacteria Settling out of the Air in Operating Roms Determined by Exposure of Culture Plates

Date 1956	O.R.	Numbers of organisms settling per hour											
		Theatre Unoccupied (Overnight)	Before First Case	During First Case	Before Second Case	During Second Case	Befre Thid Case	During Third Case	Before Fourth Case	During Fourth Case	Before Fifth Case	During Fifth Case	After Final Case
Jan. 5	II	----	43.5	40.9	48.5	28.5							27.0
12	II	----	52.5	35.3	109.6	86.4							72.0
28	VII		23.3	29.9									56.9
Feb. 2	VII	5.0	45.7	17.4	63.5	122.7	80.2	50.0					62.9
9	III	7.9	106.7	50.6	68.3	24.0							66.7
16	V	2.6	23.6	30.0	28.6	32.5	50.2	66.0	63.0	30.0			38.2
23	VI	3.3	29.1	27.5	70.9	30.3	129.0	91.9					44.5
Mar. 1	VI	2.7	22.3	17.3	103.8	11.8	35.3	11.6					60.9
8	V	3.5	50.0	56.5	78.0	47.5	91.9	39.5					127.0
15	III	11.1	92.1	99.6	90.0	29.7	35.2	26.8					39.7
22	V	12.2	68.7	138.0	98.7	33.4							34.6
29	III	11.8	23.1	36.7	84.0	19.2	49.1	23.5	95.5	30.0	60.0	410.0	30.8
May 17	V	10.7	68.6	61.6	109.4	73.8	75.9	30.0	70.3	79.7	69.1	72.0	86.9
24	VII	18.0	96.8	37.8	80.0	72.6	99.3	32.5	100.4	58.6			90.0
29	VI	27.8	33.0	79.8	88.9	27.0	17.1	25.4	29.2	22.7			209.4
30	II	8.9	66.4	39.8	41.5	40.0	46.3	44.0	46.4	46.4	40.9	42.9	273.0

(continued on next page)

TABLE 4 B

Bacteria Settling out of the Air in Operating Rooms

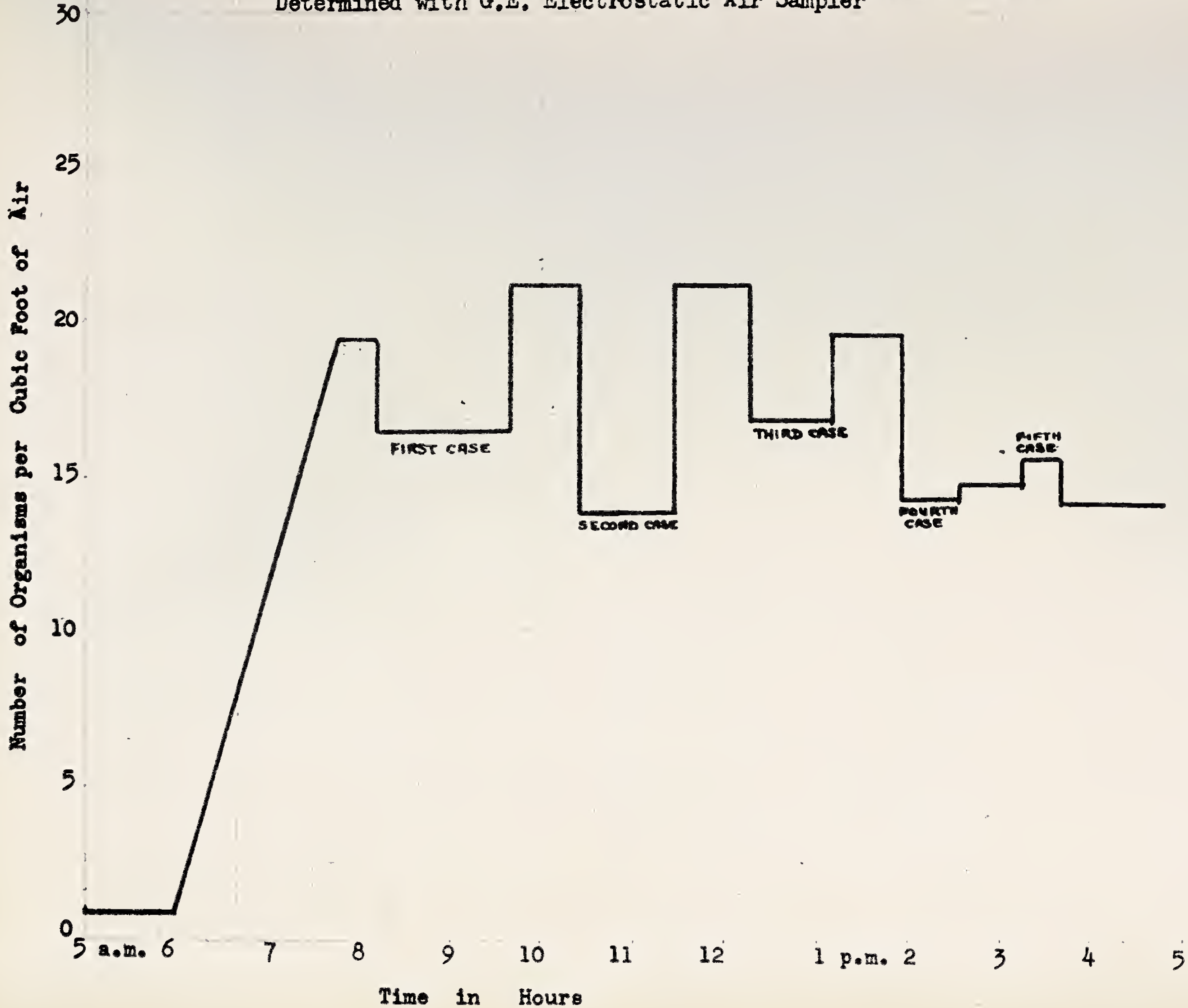
Determined by Exposure of Culture Plates

Numbers of organisms settling per hour air

Date 1956	O.R.	Theatre Unoccupied (overnight).	Before First Case	During First Case	Before Second Case	During Second Case	Before Third Case	During Third Case	Before Fourth Case	During Fourth Case	Before Fifth Case	During Fifth Case	After Final Case
June 5	I	17.2	33.8	15.7	31.1	14.5	65.5	51.4	56.8	51.7			15.0
6	I	15.9	93.5	65.8	41.5	37.5	47.1	36.1					34.8
26*	II	5.4	70.0	45.0	60.0	56.6	61.7	42.9	86.3	55.4	85.0	46.9	210.0
27	II	10.9	96.7	38.1	43.8	27.9	114.5	25.0	78.3	51.6			77.9
July 4	VI	20.7	67.1	56.4	73.6	79.3							62.0
5*	II	5.7	109.5	56.3	206.7	63.9	243.0	52.3					43.4
25*	II	23.6	62.3	32.9	96.9	59.9							66.0
Aug. 8*	II	9.1	95.6	54.0	93.7	32.8	84.2	71.4					60.0
9*	II	9.8	39.5	9.1	50.5	25.8	81.5	41.0					84.9
16	VI	17.1	60.0	9.2	63.8	37.5							49.9
17	VI	6.5	38.5	30.9									60.9
22	V	5.4	----	75.2	78.5	36.0	107.7	61.9					22.8
23	V	7.3	84.0		61.1	60.0	57.0	60.0	46.2	80.0	38.7	75.0	75.0
Sept. 6	VII	19.5	97.9	94.3	112.8	50.0	143.3	48.4	45.8	56.5			98.7
7	VII	15.3	82.5	106.7	88.2	66.2	151.7	87.9	90.0	73.8	172.0	67.5	79.5
12*	I	21.8	56.8	34.4	66.5	26.1	75.0	46.9	86.4	7.2			70.0
13*	III	27.2	101.5	63.2	114.5	76.2	140.0	88.8					59.8
25*	I	28.9	----	75.6	129.0	102.9							42.1
26*	III	8.2	80.5	32.1	57.8	75.9	63.7	28.4					58.0
Averages		12.5	64.1	49.8	80.7	48.7	85.8	47.7	68.8	49.5	77.6	119.1	74.0

* Air conditioning unit in use

General Environmental Conditions
Degree of Bacterial Contamination of Air in Operating Rooms
Determined with G.E. Electrostatic Air Sampler



Effect of Air Conditioning

Frigidaire air conditioning units were installed in operating rooms I, II and III during the period of testing. These units, consisting of forced air fans with refrigerating units and glass-wool filters, may be set to draw in outside air or to recirculate inside air. The latter procedure was followed in the University Hospital. The air conditioning units were operated during the day only. Results of air samples taken with the air conditioning units in operation are indicated by an asterisk in Tables 4 A and 4 B. These results, summarized in Table 5 A (page 32) and Figure 5 B (page 33), show increased over-all aerial contamination with air conditioning units in operation. The results indicate a continual build-up or accumulation of micro-organisms in the air of the operating theatre throughout the day when recirculation of air by the air conditioning unit is in operation.

Since outside air contains fewer pathogens than indoor air (Bourdillon, et al. 1948), it would seem advisable to set the apparatus for intake of outside air, although a more efficient filter system would be of value with the present method of operation. The ideal would be a positive pressure ventilation system with efficient filters located in air intake ducts.

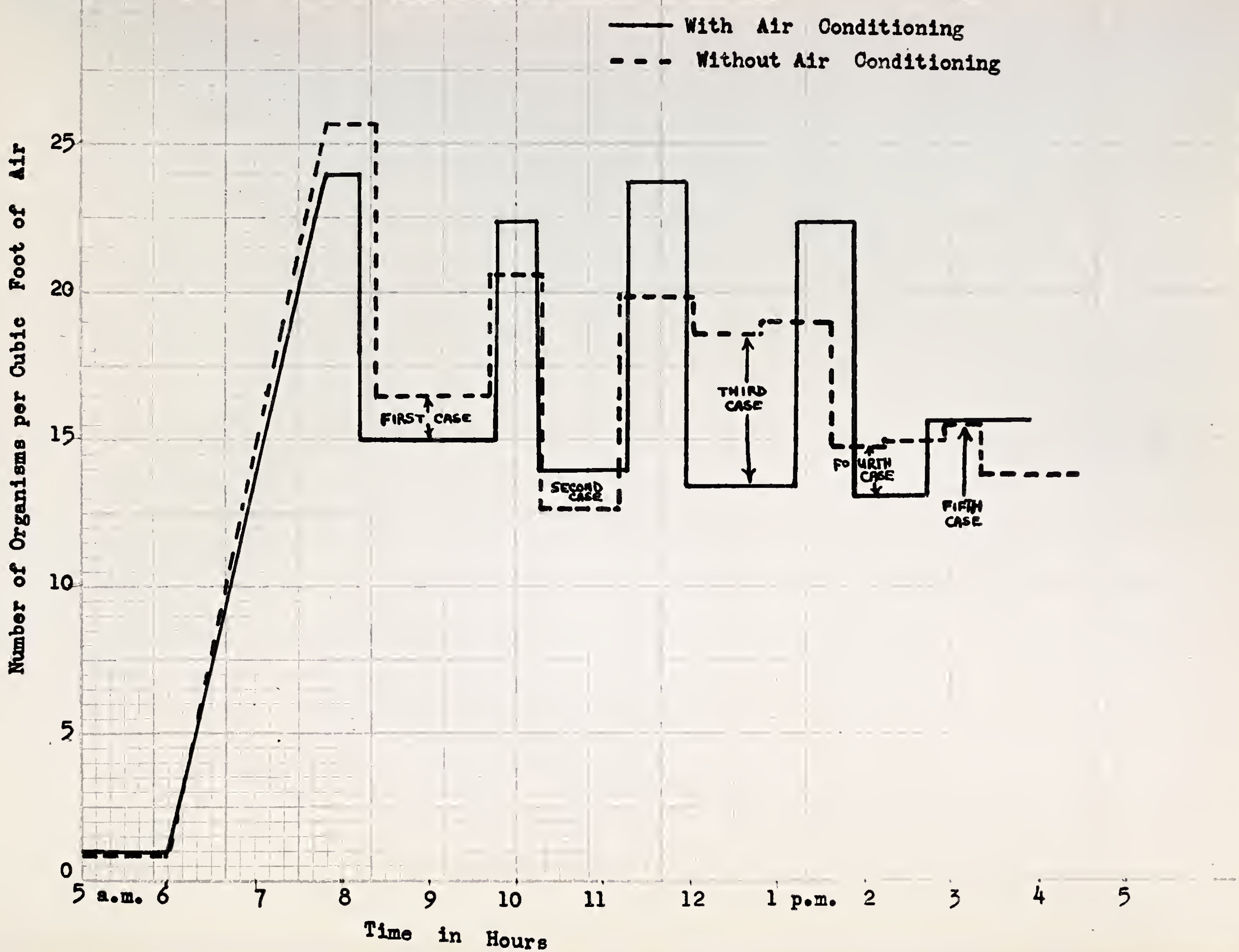
TABLE 5 A

General Environmental Conditions

Effect of Air Conditioning Units on Bacterial Contamination of Air in Operating Rooms

Time of Sampling	With Air Conditioning			Without Air Conditioning		
	No. samples taken	Average no. of organisms per cu. ft of air	Av. time	No. samples taken	Average no. of organisms per cu. ft of air	Av. time
5-6 a.m. (Theatre empty)	9	1.0	60	25	0.9	60
Before first case	9	24.1	27	25	25.6	34
During first case	10	15.0	91	26	16.4	80
Before second case	10	22.4	29	24	20.6	41
During second case	10	14.0	61	24	12.6	52
Before third case	9	23.8	39	17	19.8	53
During third case	9	13.5	68	17	18.7	47
Before fourth case	3	22.4	39	10	18.9	48
During fourth case	3	13.1	49	10	14.8	36
Before fifth case	0	----	--	5	14.9	43
During fifth case	0	----	--	5	15.6	23
After last case	10	15.7	69	25	13.7	65

General Environmental Conditions
Effect of Air Conditioning on Degree of Bacterial Contamination of Air in Operating Rooms



Effect of Traffic

Traffic was determined by counting the number of persons entering an operating theatre during each operation from the time of the incision until the time of the closure. Traffic counts are tabulated in Table 6 A (page 35). The effect of the duration of the operative procedure on traffic and aerial contamination is shown in Table 6 B (page 36) by grouping cases according to the length of time required for surgery. These results show an increased average number of entries into the operating theatre corresponding to the length of the operative procedure. No such relationship can be shown between the amount of traffic and the degree of bacterial contamination of air in the operating theatres. Traffic counts are also compared according to the type of the operative procedure in Table 6 C (page 37). These results are inconclusive because of the number of uncontrolled variables, but our observations indicate that in the case of operations of a complex or unique character the amount of traffic may greatly exceed what may be termed normal or necessary. For example, the number of entries into the operating theatre was much higher during breast biopsies than during other short operative procedures. A corresponding increase in the degree of aerial contamination is demonstrated.

TABLE 6 A

Comparison of Traffic with Duration of Operative Procedure

TABLE 6 A

Comparison of Traffic with Duration of Operative Procedure

Date 1956	O.R.	Number of entries by individuals into operating rooms					Duration of operative procedure in minutes				
		During First Case	During Second Case	During Third Case	During Fourth Case	During Fifth Case	First Case	Second Case	Third Case	Fourth Case	Fifth Case
Jan. 5	II	38	10				272	122			
12	II	55	18				182	25			
28	VII	5	22				7	58			
Feb. 2	VII	8	5	20			62	22	12		
9	III	92	7				254	25			
16	V	27	7	32	28		22	24	60	18	
23	VI	58	39	38			142	95	74		
Mar. 1	VI	21	8	20			80	22	36		
8	V	11	28	17			52	72	41		
15	III	45	35	30			138	103	94		
22	V	26	66				80	158			
29	III	11	11	54	13	00	54	25	92	26	12
May 17	V	11	15	5	17	9	38	48	34	64	25
24	VII	15	7	30	25		46	38	61	44	
29	VI	49	12	10	22		115	20	26	45	
30	II	47	8	28	00	13	86	12	39	1	14

(continued on next page)

TABLE 6 A

Comparison of Traffic with Duration of Operative Procedure

Date 1956	O.R.	Number of entries by individuals into operating rooms					Duration of operative procedure in minutes				
		During First Case	During Second Case	During Third Case	During Fourth Case	During Fifth Case	First Case	Second Case	Third Case	Fourth Case	Fifth Case
June 5	I	18	41	16	23		42	83	28	58	
6	I	20	2	19			72	8	88		
26	II	13	9	3	24	36	24	18	14	26	92
27	II	14	17	13	14		52	43	48	50	
July 4	VI	10	4				50	28			
5	II	55	49	77			114	124	148		
25	II	78	80				132	148			
Aug. 8	II	45	25	8			150	53	21		
9	II	13	36	57			87	86	136		
16	VI	13	3				59	8			
17	VI	10					68				
22	V	27	23	15			75	65	31		
23	V	5	11	4	6	7	14	22	10	12	24
Sept. 6	VII	8	30	12	4		14	96	38	17	
7	VII	3	24	17	20	25	9	68	28	48	40
12	I	16	35	28	0		61	122	64	5	
13	III	26	43	78			76	100	96		
25	I	80	14				204	21			
26	III	29	48	13			99	124	55		
Averages		28.3	23.1	25.8	14.9	15.0	86.6	61.4	55.0	31.8	34.5

TABLE 6 B

Effect of Duration of the Operative Procedure on

Traffic and the Degree of Bacterial Contamination of Air in Operating Rooms

Length of Operation	No. of Cases	Average No. of entries into O.R. during operation	Average length of operation in minutes	Average No. of organisms per cu. ft of air
0 - ½ hr.	38	9.5	18.1	16.2
½ - 1 hr.	28	16.9	45.7	12.8
1 - 1½ hrs.	20	24.9	73.6	11.6
1½ - 2 hrs.	11	43.5	99.6	14.3
2 - 2½ hrs.	11	52.9	135.1	13.6
2½ - 3 hrs.	1	66.0	158.0	12.8
over 3 hrs.	4	66.3	228.0	14.5

TABLE 6 C

Effect of Entries Made by Individuals Into Operating Rooms on Aerial Contamination

Procedure	No. Cases	Average no. of entries into O.R. during operation	Average length of operation in minutes	Average no. of organisms per cu. ft of air
Gastrectomy	4	48	188	16.6
Hysterectomy	7	46	108	15.8
Hemorroidectomy	3	15	62	9.1
Exn. cataract	6	13	57	10.3
Breast biopsy	4	44	19	14.5
Cholecystectomy	4	34	97	10.3
Hernia repair	11	28	74	13.2
T. & A.	6	11	32	12.8
D. & C.	6	7	38	12.4
Misc. eye cases	2	29	32	9.8
Misc. nasal cases	3	7	71	17.0

[illegible]

[illegible]

[illegible]

[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
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[illegible]	[illegible]	[illegible]	[illegible]	[illegible]

Effect of Temperature

Indoor temperatures were recorded at the end of each sampling time using a Taylor "Humidiguide". Outdoor temperatures were obtained from the Meteorological Division of the Department of Transport. All temperatures were reported in degrees Fahrenheit. Because of the number of variables encountered, the first case of the operating day or the first hour of the first operation for cases lasting over one hour was chosen as the test period. The activity at this period was most uniform since the major cases for the operating day were scheduled first.

Indoor and outdoor temperatures are compared with the degree of bacterial contamination of air in Table 7 A (page 39). The results are graphically presented in Figures 7 B (page 40) and 7 C (page 41).

Although the results show considerable variability, the maximum degree of aerial contamination occurred at indoor temperatures between 65 and 75 degrees Fahrenheit and outdoor temperatures of approximately 50 degrees Fahrenheit.

TABLE 7 A

Comparison of Indoor and Outdoor Temperatures

With the Degree of Bacterial Contamination of Air in Operating Rooms

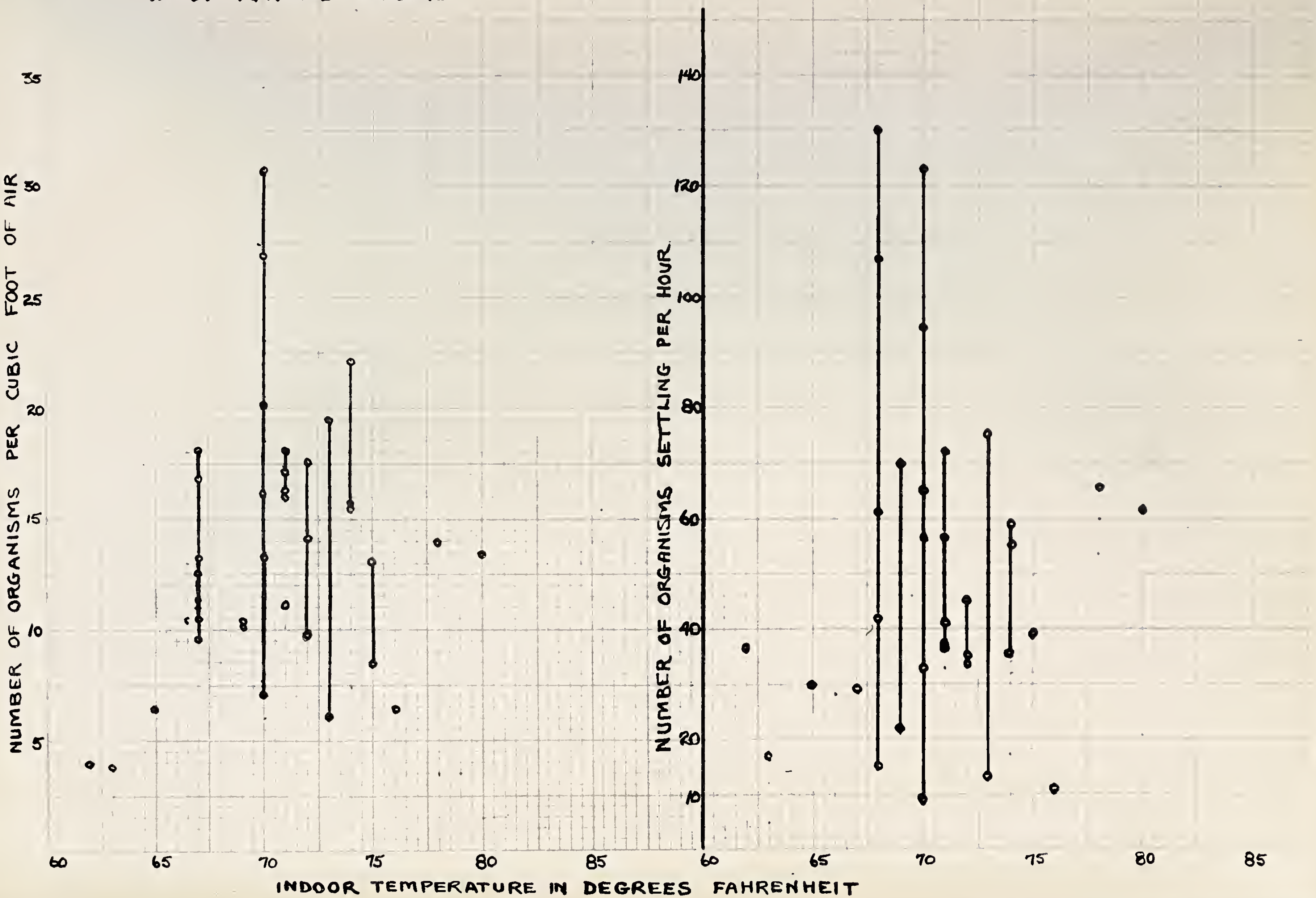
TABLE 7 A

Comparison of Indoor and Outdoor Temperatures

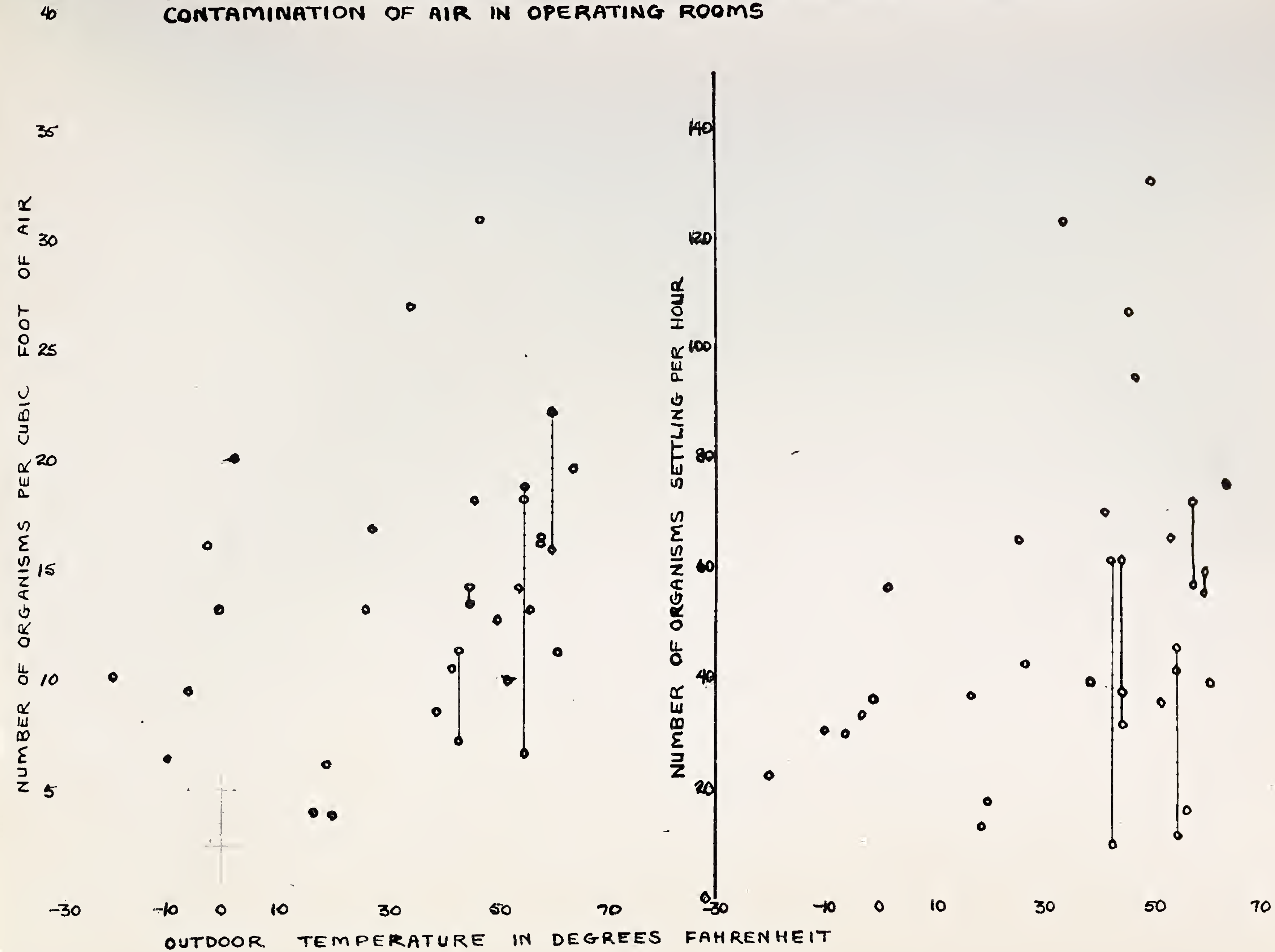
With the Degree of Bacterial Contamination of Air in Operating Rooms

Date 1956	Sampling Time in Minutes	Indoor Temperatures in Degrees Fahrenheit	Outdoor Temperatures in Degrees Fahrenheit	No. of Organisms per cu.ft of air	No. of Organisms settling per hour
Mar. 29	54	62	17	4.0	36.7
Feb. 2	62	63	20	3.9	17.4
16	22	65	-10	6.4	30.0
Jan. 28	7	67	- 6	9.5	29.9
Mar. 22	60	68	27	16.8	42.0
May 29	60	68	50	12.6	130.0
June 5	42	68	57	13.1	15.7
Sept. 7	9	68	46	18.2	106.7
113	60	68	43	11.3	61.0
25	60	69	42	10.4	70.0
Feb. 23	60	69	-20	10.2	22.0
Jan. 5	60	70	- 3	16.2	33.0
Feb. 9	60	70	26	13.3	65.0
Mar. 8	52	70	2	20.1	56.5
15	60	70	34	26.9	123.0
Aug. 16	59	70	43	7.2	9.2
Sept. 6	14	70	47	30.7	94.3
May 24	46	71	45	18.1	37.8
30	60	71	55	17.1	41.0
June 27	52	71	61	11.3	38.1
July 4	50	71	58	16.3	56.4
5	60	71	58	16.4	72.0
June 26	24	72	55	17.5	45.0
Aug. 17	68	72	45	14.2	30.9
Sept. 12	61	72	52	9.8	35.0
Aug. 22	75	73	64	19.5	75.2
Mar. 1	60	73	19	6.2	13.0
July 25	60	74	60	15.7	55.0
Aug. 8	60	74	60	22.2	59.0
Jan. 12	60	75	- 1	13.3	36.0
Aug. 23	14	75	60	----	----
Sept. 26	60	75	39	8.4	39.0
Aug. 9	60	76	55	6.5	11.0
June 6	72	78	54	14.1	65.8
May 17	38	80	45	13.4	61.6

COMPARISON OF TEMPERATURE WITH DEGREE OF CONTAMINATION OF AIR
IN OPERATING ROOMS



COMPARISON OF OUTDOOR TEMPERATURE WITH DEGREE OF BACTERIAL CONTAMINATION OF AIR IN OPERATING ROOMS



Effect of Relative Humidity

Relative humidity was recorded at the end of each sampling time using a Taylor "Humidiguide" gauge. The readings were corrected using a sling hydrometer and also by the procedure of Lidwell and Lowbury (1950). (See Appendix A.)

The first case or the first hour of the first case was chosen as a standard testing period for determining the effect of relative humidity on bacterial contamination of operating room air. Table 8 C (page 43) lists relative humidity readings, which were corrected by use of the graph in Figure 8 B (see Appendix A), and the degree of aerial contamination. The results, presented graphically in Figure 8 D (page 44), indicate that no correlation could be demonstrated between relative humidity and the degree of aerial contamination.

TABLE 8 C

Comparison of Relative Humidity with Bacterial Contamination of Air in Operating Rooms

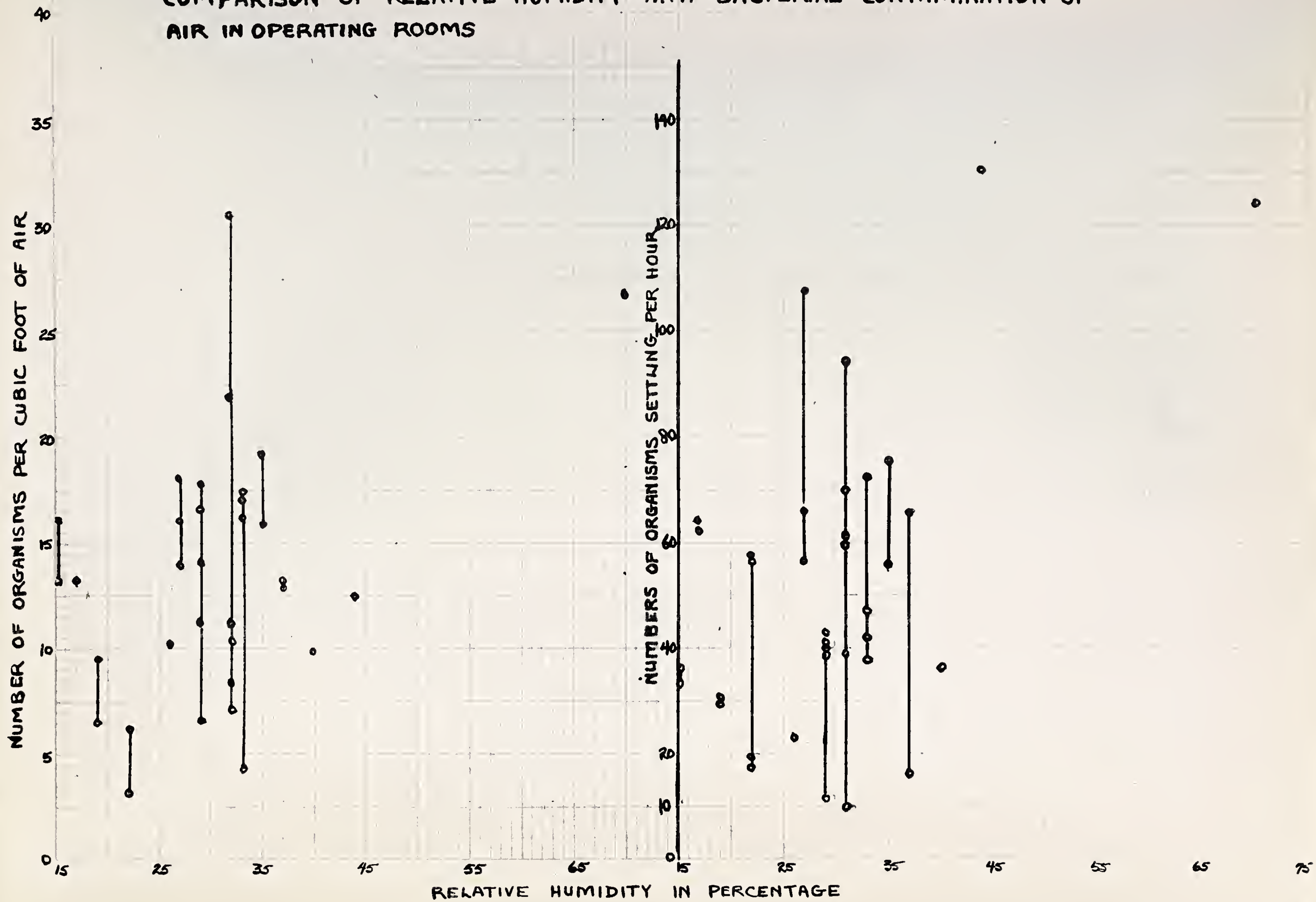
TABLE 8 C

Comparison of Relative Humidity with Bacterial Contamination of Air in Operating Rooms

Date 1956	Corrected Relative Humidity in Percentage	Sampling Time in Minutes	No. of Organisms per cu. ft of air	No. of Organisms Settling per hour
Jan. 12	15	60	13.3	36.0
5	15	60	16.2	33.0
May 17	17	38	13.4	61.1
Jan. 28	19	7	9.5	29.9
Feb. 16	19	22	6.4	30.0
2	22	62	3.9	17.4
Mar. 1	22	60	6.2	13.0
8	22	52	20.1	56.5
Feb. 23	26	60	10.2	22.0
June 6	27	60	14.1	65.8
July 4	27	50	16.3	56.4
Sept. 7	27	9	18.2	106.7
Mar. 22	29	60	16.8	42.0
May 24	29	46	18.1	37.8
June 27	29	52	11.3	38.1
Aug. 9	29	60	6.5	11.0
17	29	68	14.2	30.9
8	31	60	22.2	59.0
16	31	59	7.2	9.2
Sept. 6	31	14	30.7	94.3
13	31	60	11.3	61.0
25	31	60	10.4	70.0
26	31	60	8.4	39.0
Mar. 29	33	54	4.0	36.7
May 30	33	60	17.1	41.0
June 26	33	24	17.5	45.0
July 5	33	60	16.4	72.0
25	35	32	15.7	55.0
Aug. 22	35	75	19.5	75.2
23	35	14	----	----
Feb. 9	37	60	13.3	65.0
June 5	37	42	13.1	15.7
Sept. 12	40	61	9.8	35.0
May 29	44	60	12.6	130.0
Mar. 15	70	60	26.9	123.0

FIGURE 8D

COMPARISON OF RELATIVE HUMIDITY WITH BACTERIAL CONTAMINATION OF AIR IN OPERATING ROOMS



Summary of Results

(1) Optimum sampling volume for the General Electric Bacterial Air-sampler, determined by preliminary experiments in M 86, was 30 cu. ft. Average counts were 1.48 organisms per cu. ft with the lab. empty, 9.70 per cu. ft with students present, 9.98 per cu. ft with students present during half of the sampling period, and 2.36 per cu. ft after the students had left.

(2) Preliminary samples in operating rooms showed average counts of 0.82 per cu. ft in the unoccupied theatre and 16.67 per cu. ft in the occupied theatre.

(3) Ten series of air samples taken in various operating rooms showed average counts of 1.01 per cu. ft in the unoccupied theatre, 17.1 per cu. ft during the first case and 13.73 per cu. ft after the first case. The number of organisms settling from the air, determined by exposure of blood agar plates, was 14.1 per hour before the first case, 52.3 per hour during the first case and 45.5 per hour after the first case.

(4) Five series of air samples taken at the centre of the corridor showed counts of 26 per cu. ft at 5:00 to 6:00 a.m., 32.9 per cu. ft at 9:00 to 10:00 a.m. and 24.3 per cu. ft at approximately 11:30 a.m. to 12:30 p.m.

Another set of samples taken from the end of the corridor at the same sampling times showed counts of 1.27, 16.28 and 18.64 per cu. ft of air.

(5) Control air samples obtained in M 86 showed counts varying from 2.3 per cu. ft in the unoccupied laboratory to 14.37 per cu. ft

with students present. Air samples from M 71, a smaller room, showed counts varying from 1.47 per cu. ft in the empty laboratory to 3.9 per cu. ft with students present.

(6) A series of air samples taken during 105 operations on 35 operating days showed an average count of 0.9 organisms per cu. ft of air in unoccupied theatres, 19.5 before the first case, 13.8 to 16.8 during cases, 14.9 to 21.2 between cases, and 15.6 organisms per cu. ft after the last case.

(7) Air conditioning units resulted in a gradual increase in aerial contamination throughout the day. The increase was particularly evident during cleaning of the operating rooms between cases, probably because particulate matter disturbed at that time is prevented from "settling out".

(8) The effect of traffic was inconclusive, but there seemed to be a suggestion that traffic was influential in increasing aerial contamination.

(9) The maximum degree of aerial contamination occurred between indoor temperatures of 65 and 75 degrees Fahrenheit and outdoor temperatures of approximately 50 degrees Fahrenheit.

(10) No relationship between relative humidity and aerial contamination could be demonstrated.

(11) M. pyogenes var. aureus was contained in 67% of the 305 sets of samples taken in operating theatres. Streptococcus viridans was contained in 88% of the samples and Streptococcus pyogenes was contained in 2.5%. The number of hemolytic organisms was greatest during cleaning when masks were not worn.

(12) A total of 210 cultures of M. pyogenes var. aureus were phage typed. 107 of these were not typable. Ten were not lysed by individual phages, but showed lysis with bacteriophage pools C1 and C2.* Thirteen cultures were lysed by the phage pattern 47/Pool C. Eleven were lysed by phage type 3A and 9 cultures were lysed by phage type 81. Six cultures were lysed by each of the following phage patterns: 3A/Pool B, 7/53/70, 52/52A/81. Four cultures were lysed by bacteriophage type 53 and 4 by the phage pattern 52/52A. Three cultures were lysed by phage pattern 29/52 and 2 cultures were lysed by each of the following patterns or types: 3A/3B/3C/55, 29, 47/81/ Pool C1/C2, 6/47/81/Pool C/A, 52/52A/Pool A/B. Each of the following phage types or patterns lysed one of the cultures: 3A/47/Pool C, 3A/47/81/Pool C, 3B/55, 3A/7/Pool B, 3A/3B/3C/7/47/54/70/73/75, 6/7, 6/7/47/53/54/70/75, 7/47/53/54/70/75/77+, 7/53/54, 29/73/75, 7/47/53, 29/47/Pool C1/C2, 7/53/54/70, 52/81, 52/52A/81/Pool C, 52/52A/Pool A/B, 52/55/6/7/47/53/54/70/75+, 53/54, 53/75, 54, 55/Pool A/C, 81/Pool A/C.

*

N.T. --- non-typable

Pool A - bacteriophages 29/52/52A/79

Pool B - bacteriophages 3B/3C/55

Pool C - bacteriophages 7/42E/53/54/70/73/75/77

Pool C1 bacteriophages 6/7/53/54

Pool C2 bacteriophages 42E/70/73/75/77

+ denotes weak lysis (less than 50 plaques)

SPECIFIC ENVIRONMENTAL FACTORS

INFLUENCED BY THE DURATION OF THE OPERATIVE PROCEDURE

(b) Masking of Personnel

Historical Introduction

Masking of operating room personnel has been under discussion since first suggested by Berger in 1889. Numerous workers including Meleney (1935), Hare and Willets (1941), Davis (1934), Blair (1948) and Walter (1952) recognized the importance of masking for controlling contamination from respiratory tracts. However, the ideal construction and relative efficiency of masks are still unsettled problems. Mason Leete (1919) stated that 10 to 12 layer muslin or gauze masks are effective in preventing the passage of air-borne droplets containing *Staphylococcus aureus*. Kellogg and MacMillan (1920) used coughed, sprayed and atomized droplets to show that gauze masks which are not too thick for comfortable breathing are ineffective in preventing the passage of micro-organisms. Walker (1930) suggested the use of gauze masks with rubber deflectors in the centre. Walters (1936) recommended masks made entirely of an impervious material called "plastocele" with a cotton filter for trapping expired droplets. Arnold (1938) suggested the use of cellucotton instead of gauze for masking. Rooks et al. (1941) reviewed the efficiency of both cellucotton and gauze masks. They stated that both types of masks are efficient if thick enough and that laundering increases the efficiency of gauze masks. Abramson (1944) stated that both deflector and absorption-type masks were efficient in reducing cross-infection. Shapiro (1950) claimed that 3-layer gauze masks are sufficient to prevent cross-infection with *Mycobacterium tuberculosis*. Bourdillon et al. (1948) studied the efficiency of

various materials used for the construction of masks, including plastics, vegetable fibres, cotton fibres, gauze and cellophane. They stated that 8 layers of gauze are ineffective if the mask fits loosely, while even 2 layer gauze masks are efficient in filtering out expired organisms if the mask fits properly.

Masking at the University Hospital

Curity masks consisting of 2 layers of gauze with a heavy layer of flannelette between are used in the operating rooms. These masks are approximately 6 inches square and have tape ties at the tops and bottoms. The masks are autoclaved at a pressure of 15 pounds per square inch for 45 minutes. They are then placed in open receptacles in the hall or the surgeons' dressing room by an orderly. All operating personnel are required to wear the masks, although there is no apparent control over the length of time or manner in which the masks are worn. Persons are frequently seen entering operating theatres unmasked. Masks are not worn during cleaning of the operating theatres.

Materials and Methods

Preliminary Experiments

Cotton cloth discs approximately 1/4 inch in diameter and approximately 0.05 square inches in area were sewn on the inside and outside gauze layers at the approximate centres of the masks. The masks were then wrapped and sterilized at a pressure of 15 pounds per square inch for 20 minutes. The masks were distributed to operating personnel including

nurses, surgeons and anaesthetists to be worn instead of the regular masks. After use the masks were collected in sterile paper bags. In the laboratory the cloth discs were removed using sterile scissors, and were placed in 3 ml. volumes of nutrient broth. After incubation for 24 hours at 37.5°C., 0.1 ml. volumes of broth were spread on blood agar plates. The plates were then incubated for 48 hours at 37.5°C. before counting. The results from this first series of qualitative experiments to determine whether contamination could be detected are given in Table 9 A (page 51).

Test Series

The next series of samples were quantitative to determine the relative degree of contamination on the inside and outside of masks. After removal from the masks the discs were shaken in 10 ml. sterile water blanks until the fibres separated (at least 5 minutes). 0.1 ml. volumes were then plated on blood agar plates. All cultures were incubated 48 hours at 37.5°C. before counting colonies. Counts were multiplied by the dilution factor of 100 to determine the number of organisms per disc (0.05 square inches). The results are summarized in Table 9 B (page 52). Organisms recovered from masks included the following: diphtheroids, Neisseria, yeasts, aerobic spore bearers, Streptococcus viridans, Streptococcus pyogenes, Micrococcus pyogenes var. aureus and albus.

Controls

Masks were also sampled from the supply receptacles in the hall and

surgeons' dressing room at the beginning and end of the operating day. These were cultured in the same way except that circular areas of approximately 1/4 inch in diameter were cut from the centre of the top and bottom layers of gauze. The results of these samples are also summarized in Table 9 B (page 52).

Summary of Results

(1) Preliminary experiments showed that micro-organisms could be recovered from discs sewn on the inside and outside of masks worn by operating theatre personnel. The degree of contamination was usually less on the outside of the masks.

Contamination was demonstrated on masks sampled before use.

(2) Masks sampled after use showed an average of 11,173 organisms per disc (0.05 square inches) on the inside and 454.5 on the outside of the masks. Respiratory organisms (*Streptococcus viridans* and *Micrococcus pyogenes var. aureus*) passed through the masks on only 3 occasions. On one occasion the mask was moistened with mucus.

(3) Masks sampled before use were frequently heavily contaminated, with an average count of 88 and 733 organisms per 0.05 square inches of mask surface on the inside and outside respectively.

(4) Eleven cultures of *M. pyogenes var. aureus* isolated from the inside of masks were phage typed. Eight of these were non-typable, 1 was lysed by phage type 81 and 2 were lysed by the phage pattern 3A/Pool B.

One culture of *M. pyogenes var. aureus* isolated from the outside of a mask was non-typable.

TABLE 9 A

Degree of Bacterial Contamination of Masks Worn by Operating Personnel

Number of organisms per 0.1 ml. of nutrient broth used to wash discs sewn on inside and outside of masks after 24 hours incubation.

Sample Number	Inside	Outside
1	Confluent growth	0
2	" "	1
3	" "	0
4	" "	3
5	" "	Confluent growth
6 (control sampled from supply receptacle)	6	Confluent growth

TABLE 9 B

Bacterial Contamination of Masks Worn by Operating Theatre Personnel

	Masks Worn in Operating Theatres		Masks Sampled from Supply Receptacles in the Hall	
	Inside	Outside	Inside	Outside
Number of Masks Sampled	150	149	43	45
No. of samples with growth TNTC*	11	4	2	3
No. of samples with *M. pyogenes var. aureus*	11	1	2	0
No. of samples with *Streptococcus viridans*	26	2	0	2
No. of samples with *Streptococcus pyogenes*	1	0	0	0
Average number of organisms per disc (0.05 square inches)	11,172	454	88	733

* Too numerous to count

(c) Splash Basins

Introduction

Splash basins, i.e. basins containing sterile distilled water used to rinse instruments, sponges and surgeons' hands, have received very little mention as a possible method of conveying infection. Walter (1952) stressed the importance of adequate sterilization of distilled water used in operating rooms. Colebeck (1956) and Starkey (1956) discussed the importance of fomites in the transmission of hospital infections, but did not refer particularly to splash basins. Poppe (1943) discussed wash basin contamination with respect to the use of zephiran chloride for controlling such contamination.

Splash Basins in Operating Theatres at University of Alberta Hospital

Stainless steel basins are used in operating theatres. These are sterilized in a steam pressure sterilizer the evening before operating. Basins are usually uncovered and filled with hot or cold sterile distilled water or normal saline immediately after cleaning of the operating theatre is completed. The air in the operating theatre is still heavily contaminated at this time. The water may remain unchanged for the duration of the operative procedure or may be changed periodically. One or two basins are usually reserved for rinsing instruments or sponges which may be re-used. The remaining basins are used by surgeons for rinsing their gloves.

Materials and Methods

Test Series

One ml. samples of water were obtained usually at one-half hour intervals from splash basins using sterile 1 ml. pipettes and rubber bulbs. Samples were immediately added to 9 ml. volumes of Letheen broth (see Appendix B) which inhibits the action of quaternary ammonium compounds and supports the growth of a wide variety of micro-organisms. Samples were transferred to the laboratory as soon as possible and stored in a refrigerator at 5°C. until the necessary medium had been melted (usually ½ hour). A series dilutions of the water samples (1/10 to 1/1000) were then prepared using sterile 9 ml. water blanks. Pour plates were prepared using 1 ml. portions of each dilution with nutrient agar solids melted and cooled to 42°C. in a water bath. Cultures were also prepared using 6 drops of samples measured by calibrated pipettes, by the Reed drop-plate method (Reed and Reed, 1948; Donald, 1913,1916). Blood agar plates were used with the Reed drop-plate method to facilitate recognition of the organisms. All cultures were incubated at 37.5°C. for 48 hours before counting colonies. Plates with 7-300 colonies were chosen for counting. The minimum number of colonies counted was lower than that recommended by the American Public Health Association Standard Methods for Examination of Water & Sewage (1946), but the expected degree of contamination was of a lower order than that usually expected in waters. Plates with too few colonies and conflicting results in the various dilutions were recorded as 0. The presence of zephiran chloride, from instruments soaked in zephiran chloride solution before rinsing, may have been responsible for some of the erratic results. The use of Letheen broth, containing

a phospholipid as diluting agent may have corrected this error, although Baker et al. (1941) stated that a phospholipid must be added before or with the quaternary ammonium compound to exert a protective action against the antibacterial effect of the surface active agent. The results are given in Table 10 A (page 56).

Ten of the basins showed progressive contamination. The results for these basins are given in Table 10 B (page 57).

Summary of Results

(1) 103 of 122 basins remained sterile or the number of organisms present was too few to count throughout the operative procedure.

(2) The remaining 19 basins were contaminated. Ten of these showed progressive contamination.

(3) Fourteen basins showed slight contamination with a final average of 146 organisms per ml. Three basins had an average of 28,333 organisms per ml. Cultures of 2 of the basins had too many colonies to count in the dilutions used.

(4) The following organisms were recovered: aerobic spore bearers, diphtheroids, coliforms, Actinomycetes, moulds, Gaffkya and Micrococci including *Micrococcus pyogenes var. aureus* and *albus*.

(5) *Micrococcus pyogenes var. aureus* was recovered from 11 of the basins.

(6) All cultures of *M. pyogenes var. aureus* were phage typed. Eight were non-typable, while two were not lysed by individual bacteriophages but showed lysis with phage pools C1 and C2. The remaining culture was lysed by phages 29/Pool C1.

TABLE 10 A

Degree of Bacterial Contamination of Splash Basins

TABLE 10 A

Degree of Bacterial Contamination of Splash Basins

Date 1956	Case No.	Approximate Length of Exposure in Minutes	Number of organisms per ml.		
			Basin A	Basin B	Basin C
Jan. 12	1	285	450	0	
28	2	150	0		
Feb. 2	1	120	0		
	2	120	0	0	0
Feb. 9	1	270	150	0	0
23	1	150	TNTC*	0	0
	2	120	0	0	
	3	75	0	64,000	
Mar. 8	1	75	0	0	
	2	90	0		
	3	60	0		
15	1	105	0	0	
	2	135	0		
	3	90	0		
22	1	105	0	0	
	2	135	0	0	
29	1	90	90		
	2	135	0		
	3	105	TNTC*	90	
	4	60	0		
	5	50	0		
May 17	1	60	100		
	2	60	9,000		
	3	40	12,000	0	
	4	160	390		
24	1	15	0		
	2	90	0		
	3	105	0		
	4	60	0		
29	1	120	0	0	
	3	60	0		

* Too numerous to count

TABLE 10 A

Degree of Bacterial Contamination of Splash Basins

	3	75	0		
	5	30	90	0	
June 5	1	90	170		
	2	135	0	0	
	3	120	0		
	4	60	70	0	0
6	1	105	0		
	3	75	0	0	
26	1	60	0		
	2	30	0		
	3	30	0		
	4	120	0	0	
27	1	60	0		
	2	45	0		
	4	45	0	80	
July 4	1	60	0		
	2	60	0		
5	1	105	0		
	2	120	70		
	3	180	100		
25	1	165	0	70	0
	2	150	0		
Aug. 8	1	165	0		
	2	150	0		
9	1	105	0		
	2	105	0		
	3	210	0	0	
16	1	90	0		
17	1	90	0		
22	2	10	0		
	5	60	0		
23	1	10	0		
	2	120	0		
	3	50	0		
Sept. 6	2	180	0	0	
	3	60	0		
	4	45	0		
7	2	105	0		
	3	30	0		
	4	75	0	0	0
	5	60	0		
12	1	75	0	0	
	2	135	130		
	3	60	0	0	
	4	30	0		
13	1	105	0	0	
	2	120	0	0	
	3	120	0		
25	1	165	0	0	0
	2	45	0	0	
26	1	105	0		
	2	105	0	0	
	3	10	0	0	
	Number of Samples:		85	31	6
	Average Counts:		275	2072	0

Final Average Number of Organisms per ml.: 725

TABLE 10 B

Comparison of Length of Exposure

With Degree of Bacterial Contamination of Splash Basins

Date	Basin Number	Number of organisms per ml. Length of exposure in minutes				
		0 - 30	30 - 60	60 - 120	120 - 180	180+
Jan. 12	1 A	300		380	450	
Feb. 9	1 A	0	0	20	20	150
23	1 A	110		TNTC*	TNTC*	
May. 17	1 A	20	100			
	2 A	80	9000			
	3 A	8000	12000			
June 5	1 A	10	30	170		
July 5	2 A	30	20	70		
	3 A	50	10	30	100	
Sept.12	2 A	10	0	10	130	

TNTC* -- Too numerous to count in 1/100 dilution

(d) Operating Tables

Introduction

Historical

Numerous workers cite the importance of fomites in the transfer of hospital infections, although operating tables have not been mentioned specifically.

In regard to surgical linen, Beck and Colette (1952) stated that draping materials are effective for preventing the transmission of micro-organisms when dry, but allow the passage of micro-organisms when wet. Corry (1950) suggested the use of 2% lysol soaks to prevent contamination of materials, while Propst (1953) recommended the use of a solution containing hexachlorophene. Baker and Madden (1955) recommended the use of diaperine chloride 1/500 solution to prevent the passage of contamination through moist materials. Various agents have been suggested for use as disinfectant barriers in dressings, including octyl cresol compounds and other phenyl mercuric salts (Lowbury and Hood, 1952). Beck and Colette (1952) suggested the possible use of a waterproof and porous material for surgical linen to provide a mechanical barrier against the transfer of micro-organisms. Their criteria would probably be satisfied by the nylon material used by Schilling et al. (1950) for dressings, although the cost would be a prohibiting factor.

Operating Tables at the University of Alberta Hospital

Stainless steel operating tables are used in all operating theatres at the University Hospital. Mats used for operating tables are of foam rubber with a heavy plastic covering. Operating tables and mats are

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[illegible]

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[illegible]

[illegible]

[illegible]

[illegible]

washed with an aqueous solution of zephiran chloride (1/1000) at the end of each operating day, after infected cases or when grossly contaminated. Mats are covered with freshly laundered non-sterile sheets which are changed after each case. No effort is made to control dust arising from sheets and sheets are not chemically treated for this purpose. Contaminated laundry is not separated for treatment.

Materials and Methods

Operating Table Mats and Sheets

A sterile aluminum template with an area of 1 square inch cut out of the centre was used to limit the area of operating table mat which was swabbed before and/or after cases. Samples of sheets were later obtained by the same procedure. Swabs were moistened in nutrient broth before swabbing. In the laboratory swabs were shaken manually or mechanically in 3 ml. volumes of nutrient broth for 15 minutes. 0.1 ml. volumes of broth were then plated on blood agar plates. Cultures were incubated aerobically at 37.5°C. for 24 hours before counting colonies. This sampling procedure was used during 93 cases. The results of cultures of mats and sheets are shown in Tables 11 A and 11 B (pages 60, 61, 62, 63 and 64). Results are listed as the number of organisms per square inch of table surface and were obtained by multiplying the counts by the dilution factor of 30. The results are summarized in Table 11 C (page 65).

TABLE 11 A

Degree of Bacterial Contamination of Operating Table Mats

Number of Organisms per Square Inch of Mat Surface

	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
Date 1956	Before	After	Before	After	Before	After	Before	After	Before	After
Jan. 12	30	15								
28	0	0								
Feb. 2	0		0							
9	90	740	0							
16			180	0						
23					960	290				
Mar. 1	96		TNTC*		0	0				
8	0		0		60					
15					210	120				
22				30						
29			180		150		30		150	30
May 17	300	0	0	0	60	0	60	60	60	90
24	180	120	180	30	90	120	30	120		
29		30	30	30	60	60	0	0		
30		30	60	30	30	30	0	90		

[illegible]

TABLE 11 A (continued)

Date 1956	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
	Before	After	Before	After	Before	After	Before	After	Before	After
June 5	120	30	60	TNTC*	60	0		60		
6	270	270	120	180	120					
26		60	210	30		30	30	90		
27	0	30	150	0	1680	120		30		
July 4	60	0	0	660						
5		0	120	120	30	60				
25	30	30	0	90						
Aug. 5	60	30	0	0	0	0				
9		60	0	30	0	30				
16		30		0						
17		30								
22		0	0	0	0	240				
23			390	0	0	0	0	0	0	30
Sept. 6	2130	90	30	90	60	0	60	0		
7	60	TNTC*	30	30	30	30		0	450	30

TABLE 11 A (continued)

Date 1956	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
	Before	After	Before	After	Before	After	Before	After	Before	After
Sept. 12		30	30	0	30	0	60		60	30
13	0	30	30	120		30				
25	90	0	30	120						
26		0	0	0	0	60				
No. of Samples	18	25	28	24	20	20	9	10	5	5
Average No. of Organisms per Square Inch	195	63	70	64	175	61	30	45	144	42

[illegible]

TABLE 11 B

Degree of Bacterial Contamination of Operating Table Sheets

Number of Organisms per Square Inch of Sheet Surface

Date 1956	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
	Before	After	Before	After	Before	After	Before	After	Before	After
Mar. 22	30		TNTC*	45						
May 17	120	30	30		120	30	0	0	30	30
24	90	120	0	30	30	2010	0	60		
29		30	210		270	30	150			
30		90	30		60	0			240	90
June 5	0		90	0	30	0	0	90		
6	450	180	30	90						
26		0	360	120			0	30		
27	0		0		30	120		90		
July 4	30	0	0	0						
5			30		120	0				
25	0	150	0	120						
Aug. 8	90	840	60	0	60	60				
9	3930		90	30	0	0				

TABLE 11 B (continued)

Date 1956	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
	Before	After	Before	After	Before	After	Before	After	Before	After
Aug. 16	60	90	90	720						
17		300								
22			30							
23			0		30	30	30		30	30
Sept. 6	0	630	TNTC*	TNTC*	60	0	0	30		
7			TNTC*	0	0	0		360	0	120
12		90	0		0	30	0	0		
13	60	30	150	120		0				
25	60	30	0	420						
26		30	0	0		90				
No. of Samples	16	16	25	15	13	15	8	8	4	4
Average No. of Organisms per Square Inch	307	165	54	121	62	160	22	82	75	67

* TNTC -- Too numerous to count

TABLE 11 C

Comparison of Degree of Bacterial Contamination of Operating Table Mats and Sheets

	Mats		Sheets	
	Before Case	After Case	Before Case	After Case
Number of samples	80	84	66	58
Number of samples with *Micrococcus pyogenes var. aureus*	2	3	3	1
Number of samples with growth too numerous to count	1	2	3	1
Average number of organisms per square inch of surface	125	60	118	134

<u>Summary of Results</u>

(1) Cultures of one square inch areas of operating table, mats and sheets were obtained before and/or after 93 cases.

(2) Although usually very few micro-organisms were recovered, heavy contamination did occur occasionally.

(3) The average number of organisms per square inch of mat surface was 125 before cases and 60 after cases.

(4) The average number of organisms per square inch of sheet surface was 118 before cases and 134 after cases.

(5) A total of 3 cultures from mats and 4 from sheets showed growth too numerous to count.

(6) <u>Micrococcus pyogenes var. aureus</u> was recovered from 5 cultures of mats and 4 of sheets.

(7) All cultures of <u>M. pyogenes var. aureus</u> were phage typed. Cultures from mats were non-typable except one which was lysed by the phage pattern 81/52/52A.

One culture from sheets was lysed by each of the two phage patterns 29/52/47/Pool C1/C2 and 47/Pool C1/C2. The remaining cultures were non-typable.

FACTORS AFFECTING CLEANING

(e) Scrub Water for Operating Theatre Floors

Introduction

Historical

The primary concern of most workers in regard to contamination from floors has been the prevention of dust. The Medical Research Council War Memorandum (1944) discussed the relation of dust from sweeping and bedclothes to wound infections. Hare and Willets (1941) suggested the oiling of floors before sweeping. Clayton and Robertson (1945) and Anderson et al. (1944) recommended the use of spindle oil for floors to prevent the spread of respiratory organisms in dust. Colebrook (1955) also recommended oiling of floors. Walter (1952) suggested frequent cleaning of operating theatre floors to reduce contamination from this source. Starkey (1956) favoured either oiling or frequent cleaning of floors. He suggested mopping of operating theatre floors once or twice daily using oiled or wet mops. Spindle oil or glycerin was recommended for dust laying. Fixanol C, Roccol and Wescodyne were the agents recommended for use in scrubbing of floors.

Cleaning of Operating Theatres at the University of Alberta Hospital

All operating theatres at the University Hospital have marble floors. A solution of Germa Medica which contains 2% hexachlorophene in water was used for scrubbing floors in operating theatres. A small undetermined amount of Dettol was added to the solution. This mixture was renewed occasionally, but was usually used throughout the operating day.

Operating theatres were mopped after each case using mops soaked in the scrubbing mixture. Furniture was moved and the floors scrubbed thoroughly at the end of the day using a similar solution. The string mops were not decontaminated after use and were usually left in buckets used for the scrubbing mixture. Buckets and mops were stored along with a garbage receptacle in a small room opening directly onto the operating wing corridor. No waxing, oiling or dust laying measures were used.

Materials and Methods

Test Series

A total of 16 samples of the scrubbing mixture were obtained at various times using sterile 4 ounce water sample bottles. In the laboratory serial dilutions of the samples to 1 in 10^6 were prepared using 9 ml. sterile water blanks. Pour plates were then prepared using 1 ml. portions of the dilutions with 9 ml. nutrient agar, melted and cooled to 42°C. Duplicate plates of one or more of the greater dilutions were also prepared by the Reed drop-plate method using blood agar plates to facilitate recognition of the colonies. All cultures were incubated aerobically at 37.5°C. for 48 hours before counting colonies. Cultures of M. pyogenes var. aureus were isolated to determine their coagulase reaction and phage type. The results are given in Table 12 A (page 70). Results are grouped according to the time of sampling in Table 12 B (page 71).

Summary of Results

(1) A total of 16 samples of scrub water used for operating theatre floors was obtained. All samples showed heavy contamination with an average of 74,237,500 organisms per ml.

(2) Contamination was greatest after the first case (92 million organisms per ml.) and showed a progressive decrease throughout the day (to 45 million organisms per ml.)

(3) Organisms which grew on the culture plates were usually coliforms, although others, including Micrococci, Actinomyces and aerobic spore bearers, were sometimes present.

(4) *Micrococcus pyogenes var. aureus* was recovered from 5 of the samples. Two of these cultures were coagulase positive.

(5) Coagulase positive cultures of *M. pyogenes var. aureus* were phage typed. Both were non-typable.

[illegible]

[illegible]

[illegible]

[illegible]

[illegible]

TABLE 12 A

Degree of Bacterial Contamination

of Scrub Water for Operating Theatre Floors

Date	Case No.	No. of organisms per ml.
July 25	1	27,400,000
	2	15,200,000
Aug. 8	1	26,300,000
	2	23,300,000
9	1	22,200,000
	2	8,500,000
16	1	430,000,000
17	1	68,000,000
22	1	26,400,000
	2	7,800,000
Sept. 7	2	79,000,000
	4	45,000,000
13	1	45,400,000
	3	11,300,000
25	2	208,000,000
26	3	154,000,000

Number of Samples: 16

Average number of organisms per ml. 74,237,500

[illegible]

[illegible]

[illegible]

[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]
[illegible]		
[illegible]		[illegible]
[illegible]	[illegible]	
[illegible]		
[illegible]	[illegible]	
[illegible]		[illegible]
[illegible]		[illegible]
[illegible]	[illegible]	[illegible]
[illegible]		
[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	
[illegible]	[illegible]	[illegible]
[illegible]		
[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]
	[illegible]	
[illegible]	[illegible]	[illegible]

TABLE 12 B

Degree of Bacterial Contamination

of Scrub Water for Operating Room Floors

Case No.	No. of Samples	Average No. of organisms per ml.
1	7	92,243,000
2	6	56,967,000
3	2	82,650,000
4	1	45,000,000

Total 16

Average No. of organisms per ml. 74,237,500

[illegible]

[illegible]

[illegible]

[illegible]

[illegible]

[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]
	[illegible]	[illegible]

[illegible]

[illegible]

(f) Mops

Materials and Methods

Test Series

A total of 20 samples of 1 inch strands of mop fibre were obtained during the mopping procedure. The samples were immediately immersed in 4 ml. volumes of sterile serum broth. (Serum broth was used to inactivate any hexachlorophene which might be retained in the mop fibres.) In the laboratory after shaking the broths for 15 minutes, either manually or mechanically, a series dilutions of the samples to 1 in 10^3 were prepared using 4 ml. volumes of serum broth. Because of the degree of contamination the dilution series was later extended to 1 in 10^6. 0.1 ml. volumes of each dilution were plated on blood agar plates. All cultures were incubated aerobically at 37.5°C. for 48 hours before counting colonies. Counts were multiplied by the dilution factors to determine the number of organisms per inch of mop fibre. The results are given in Table 13 A (page 73). Because of the variation in dilutions used, cultures with too many colonies to count have the dilution listed in brackets. The results are grouped according to the time of sampling in Table 13 B (page 74).

Summary of Results

(1) Twenty-one 1 inch strands of mop fibre were sampled from mops used to scrub operating room floors. Cultures of 10 of these samples had too many colonies to count, while cultures of the remaining 11 samples showed an average of 6,528,560,000 organisms per inch of mop fibre.

(2) The degree of contamination was highest during mopping after the first case and was reduced after each succeeding case.

(3) Organisms which grew on culture plates were of the coliform type. M. pyogenes var. aureus (coagulase negative) was recovered from one sample.

TABLE 13 A

Degree of Bacterial Contamination

of Mops Used to Scrub Operating Theatres

Date 1956	Case No.	Number of Organisms per Inch of Mop Fibre
July 4	1	TNTC* $(1/10^2)$
25	1	TNTC $(1/10^3)$
	2	TNTC $(1/10^3)$
Aug. 8	1	TNTC $(1/10^3)$
	2	TNTC $(1/10^3)$
16	1	TNTC $(1/10^4)$
17	1	12,800,000
22	2	272,000,000
23	1	20,320,000,000
	2	TNTC $(1/10^6)$
Sept. 6	1	122,400,000
	2	62,400,000
	3	140,000,000
7	1	TNTC $(1/10^3)$
12	1	15,200,000,000
	2	14,720,000,000
13	1	TNTC $(1/10^6)$
	2	156,000,000
25	2	8,400,000
26	1	20,800,000,000
	2	TNTC $(1/10^6)$

No. of Samples 21
No. with growth TNTC 10
Average no. of organisms per inch of mop fibre 6,528,560,000

* Too numerous to count

[illegible]

[illegible]

[illegible]

[illegible]	[illegible]	[illegible]

TABLE 13 B

Degree of Bacterial Contamination

of Mops Used to Scrub Operating Theatre Floors

Case No.	No. of Samples	No. of Samples with growth TNTC*	Average No. of Organisms per Inch of Mop Fibre
1	11	6	11,291,200,000
2	8	3	384,360,000
3	2	1	14,000,000
Totals	21	10	

Average No. of Organisms per Inch of Mop Fibre 6,528,560,000

* Too numerous to count

[illegible]

[illegible]

[illegible]

[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]

[illegible]

[illegible]

(g) Floors

Materials and Methods

Test Series

Sterile aluminum templates were used to isolate 1 inch areas of operating theatre floor surface near the base of the operating table. The 1 square inch area of floor was swabbed before and after the mopping procedure. Swabs were moistened in sterile serum broth before swabbing and were immersed in 4 ml. volumes of serum broth immediately after. In the laboratory the swabs were shaken in the broths either manually or mechanically for 15 minutes. 0.1 m. volumes of the broths were then plated on blood agar plates. All cultures were incubated aerobically for 48 hours at 37.5°C. before counting colonies. Counts were multiplied by the dilution factor of 40 to determine the number of organisms per square inch of floor surface. The results are given in Table 14 A (page 76). Results are grouped according to the sampling period in Table 14 B (page 77). Cultures of M. pyogenes var. aureus were isolated for determining coagulase reactions and for phage typing.

[illegible]

[illegible]

[illegible]

[illegible]

TABLE 14 A

Degree of Bacterial Contamination of Operating Theatre Floors

Degree of Bacterial Contamination of Operating Theatre Floors

Number of Organisms per Square Inch of Floor Surface

Date 1956	Case No.	Before Mopping	After Mopping
July 25	1	360	TNTC*
	2	2,160	TNTC
Aug. 8	1	1,680	TNTC
	2	480	200
	3	0	
9	1	0	TNTC
	2	40	TNTC
	3	360	
16	1	40	
	2	360	TNTC
17	1	40	TNTC
22	1	200	TNTC
	2	1,040	TNTC
	3	120	
23	1	160	TNTC
	2		TNTC
	3	40	TNTC
	4	40	TNTC
	5	120	
Sept. 6	1	80	TNTC
	2	120	TNTC
	3	80	TNTC
	4	80	TNTC
7	2	80	TNTC
	3	40	TNTC
	4	40	TNTC
	5	120	
12	1	40	TNTC
	2	80	920
	3	400	TNTC
	4	680	TNTC
13	1	1,680	TNTC
	2	TNTC	
	3	520	200
	4		TNTC
25	1	80	
	2	80	TNTC
26	1	640	TNTC
	2	TNTC	
	3	TNTC	TNTC
No. of Samples		38	31
Average no. of organisms per square inch of floor surface		346	TNTC

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[illegible]

[illegible]

TABLE 14 B

Degree of Bacterial Contamination of Operating Theatre Floors

	Before Mopping			After Mopping		
Case No.	No. of Samples	No. with growth TNTC*	Average No. of organisms per square inch of floor surface	No. of Samples	No. with growth TNTC	Average No. of organisms per square inch of floor surface
1	12	0	451	11	10	TNTC
2	11	2	493	10	8	TNTC
3	9	1	200	5	5	TNTC
4	4	0	209	5	5	TNTC
5	2	0	120	0	---	-----

* Too numerous to count

Summary of Results

(1) Thirty-eight cultures of floors obtained by swabbing before mopping showed an average of 346 organisms per square inch of floor surface while 31 cultures obtained after mopping showed growth too numerous to count.

(2) Cultures obtained before mopping showed varying amounts of growth which indicated progressively decreasing contamination of floors. These results followed similar decreases in contamination of the scrub water and mops.

(3) All cultures except 3 showed that the mopping procedure resulted in an enormous increase in the degree of contamination, although most of the organisms could not be recovered before the next mopping. The 3 cultures showing a lower degree of contamination were obtained approximately one half hour after the mopping when the floor had dried.

(4) Organisms present in all cultures were of the coliform type.

(5) M. pyogenes var. aureus was recovered from 2 of the samples obtained before mopping and 4 of these obtained after mopping. Two of the cultures obtained after mopping were coagulase positive.

(6) The 2 coagulase positive cultures of M. pyogenes var. aureus were phage typed. One was non-typable and the other was lysed by phage type 3A.

FACTORS CONCERNED WITH ANAESTHESIA

(h) Anaesthesia Masks

Introduction

Historical

Although the maintenance of sterility of equipment used in anaesthesia is often overlooked, it has been shown that such equipment may be a route for the transmission of pathogenic micro-organisms. Joseph (1952) stated that anaesthesia increased the patient's susceptibility to respiratory infection. Contamination of anaesthesia equipment was highest immediately after use and decreased gradually. The organisms present included pathogenic Micrococci. Simple rinsing was not sufficient for the removal of micro-organisms. The author recommended the use of pHisohex for this purpose. The same agent was also used by McDonald et al. (1955) who demonstrated contamination of face masks, endotracheal tubes, tubes of the gas machines, etc. The workers claimed that autoclaving or cold sterilization was inadequate for anaesthesia equipment. Prenzlau and Karp (1954) advised autoclaving of anaesthesia equipment enclosed in cellophane tubes. Ziegler and Jacoby (1956) used tuberculosis patients to show that contamination was not usually transferred to gas machines, but tubes, masks and cuffs could be contaminated. They suggested periodical treatment of gas machines and treatment of the later types of equipment with 70% alcohol or boiling water immediately after use.

Use of Anaesthesia Masks at the University of Alberta Hospital

Rubber or plastic face masks are used routinely in administering inhalation anaesthetics. Masks were formerly washed only occasionally and were usually stored in the metal gas machine cabinets without any decontaminating procedure.

Materials and Methods

Test Series

Masks were cultured before use, after use or when stored in a cabinet. The entire inner surface of each mask was swabbed with a swab moistened in nutrient broth. The swab was then placed in a 3 ml. volume of nutrient broth and returned to the laboratory. In the laboratory the swabs were shaken mechanically or manually in the nutrient broths for 15 minutes. 0.1 ml. volumes of broth were then plated on blood agar plates. After 24 hours incubation the swabs were streaked on blood agar plates as a check on the first procedure. All culture plates were incubated aerobically at 37.5°C. for 48 hours before counting colonies. Counts were multiplied by the dilution factor of 30 to determine the number of organisms per mask. The results are given in Table 15 A (page 81).

TABLE 15 A

Degree of Bacterial Contamination of Anaesthesia Masks

Number of Organisms per Mask

Date 1956	Before Use	After Use	Samples from Storage Cabinet			
Feb. 2	910					
		1800				
Feb. 16			240	270	30	90
Feb. 23			120	5340	30	
March 1		36,000				
		75,000				
March 22			4810	330		
March 29			9390	690		
May 17			240	990	3030	5310
May 24			930	990		
May 29			120	390		
May 30			2970	3780		
No. of Samples	1	3	21			
Average No. of Organisms per Mask	910	37,600	1909			

[illegible]

[illegible]

[illegible]

[illegible]

Decontamination Procedure

Because masks were almost always heavily contaminated it was decided to adopt a decontamination procedure consisting of a thorough washing in Germa Medica (containing 2% hexachlorophene) and a half hour soak in aqueous Zephiran chloride solution 1/1000 followed by a rinsing in tap water. In practice the soaking period varied from a momentary dip to an overnight soaking.

In the next series of tests masks were cultured before use, after use, after the decontaminating procedure and also from the storage cabinets. The method of sampling was the same as that used before, except that nutrient broth was replaced with Letheen broth to inactivate the Zephiran chloride which might be present. Cultures of M. pyogenes var. aureus were isolated for determining coagulase reactions and phage typing. The results of cultures of masks taken before use, after use and after decontamination are compared in Table 15 B (pages 83, 84). The results of cultures of masks sampled from the storage cabinets are given in Table 15 C (page 85). The results of all cultures are summarized in Table 15 D (page 86). Organisms recovered included Neisseria, Streptococcus viridans, diphtheroids, moulds, Actinomycetes, Micrococci, aerobic spore bearers and Pseudomonas pyocyaneus.

Two preliminary cultures were taken of connecting tubes of the gas machine. One culture yielded 330 colonies of Micrococci, diphtheroids and Neisseria per swab. The other yielded confluent growth of Pseudomonas pyocyaneus.

TABLE 15 B

Effect of Decontamination on

Degree of Bacterial Contamination of Anaesthesia Masks

Number of Organisms per Mask

Date 1956	Case No.	Before Use	After Use	After Decontamination
May 30	1			30
	2			150
June 5	1			0
	2			30
	3			90
	4		TNTC*	30
June 6	1	60		
	2	360		
	3	0		
June 26	1	TNTC*		
	3		3090	
	4	TNTC	TNTC	
June 27	1		2700	
	3	4140	7020	
	4	60		
July 5	2	90	3870	0
	3	0		
July 25	1	180		
	2	90		
Aug. 8	1	60	780	
	3		1140	
Aug. 22	3	690	2400	

* Too numerous to count

(continued on page 84)

[illegible]

TABLE 15 B (continued)

Date 1956	Case No.	Before Use	After Use	After Decontamination
Aug. 23	1		TNTC	0
	5	60		
Sept. 6	1	2040		900
	4	840		780
Sept. 7	1	3780	TNTC	
	3	450		
Sept. 12	1		1950	
	2 & 3		10,800	
	4		780	0
Sept. 13	1	4620		
	2	3300		
	3		2970	
Sept. 26	1	3420	4500	
	2	4500		
	3	4020		
No. of Samples		24	18	11
Average No. of Organisms		1489	3000	183

TABLE 15 C

Degree of Bacterial Contamination of Anaesthesia Masks

Sampled From Storage Cabinets

Number of Organisms per Mask

Date 1956	Sample Number 1	2	3	4	5
July 4	30	210	0	30	60
July 5	60	TNTC*	570		
July 25	18,000	90	30	90	
Aug. 5	11,340	120	120	900	
Aug. 9	120	540	TNTC		
Aug. 16	TNTC	1680	TNTC		
Aug. 22	60	6650	240	60	
Aug. 23	60				
Sept. 6	120	60	2860	120	
Sept. 7	360	90	60		
Sept. 12	5820				
Total No. of Samples	58				
No. with Growth TNTC	4				
Average No. of Organisms per Mask	936				

* Too numerous to count

TABLE 15 D

Comparison of Degree and Types of Bacterial Contamination of Anaesthesia Masks

	Before Use	After Use	After Decon-tamination	Sampled From Cupboard
Number of samples	25	21	11	79
No. of samples with hemolytic *M. pyogenes var. aureus*	3	1	1	2
No. of samples with *Ps. pyocyaneus*	0	1	0	1
No. of samples with growth TNTC*	2	4	0	4
Average No. of organisms per mask	1403	8518	183	1209

* Too numerous to count

Summary of Results:

(1) Cultures of 21 masks sampled from the storage cabinets contained an average of 1909 organisms per mask. One mask sampled before use contained 910 organisms, while 3 masks sampled after use contained an average of 37,600 micro-organisms per mask. Micro-organisms which grew on the culture plates included diphtheroids, moulds, Actinomycetes, Neisseria, aerobic spore bearers, Micrococci and _Streptococcus viridans_. _Pseudomonas pyocyaneus_ was recovered from one of the masks sampled from the storage cabinet.

(2) After the decontamination procedure was adopted 24 masks were sampled before use. The average count was 1489 organisms per mask. Two of the cultures contained growth too numerous to count. Three of the cultures contained _M. pyogenes var. aureus_, two of which were coagulase positive.

(3) Eighteen masks sampled after use had an average of 3000 micro-organisms per mask. Twenty-four of the cultures contained growth too numerous to count. One of the cultures contained coagulase positive _M. pyogenes var. aureus_. One of the cultures contained _Pseudomonas pyocyaneus_.

(4) Eleven masks sampled after decontamination had an average count of 183 micro-organisms per mask. One of these cultures contained coagulase positive _M. pyogenes var. aureus_.

(5) Fifty-eight masks sampled from the storage cabinets had an average count of 936 micro-organisms per mask. Four of the cultures contained growth too numerous to count. Two of the cultures contained coagulase positive _M. pyogenes var. aureus_.

(6) One of 2 cultures of connecting tubes for the gas machine gave a count of 330 organisms, while the other had confluent growth of _Pseudomonas pyocyaneus_.

(7) Six coagulase positive cultures of _M. pyogenes var. aureus_ were phage typed. One showed weak lysis with phage type 52 and the remaining 5 were non-typable.

(i) Endotracheal Tubes

Introduction

Decontamination of Endotracheal Tubes at the University of Alberta Hospital

Endotracheal tubes used in anaesthesia were routinely washed in Germa Medica or pHisohex solution, soaked in an aqueous solution of Zephiran chloride, rinsed in tap water, and hung in an open cupboard until required for use.

Methods and Materials

Preliminary Experiments

Each endotracheal tube was placed in 100 ml. of sterile Letheen broth and shaken manually. 0.1 ml. volumes of broth were then plated on blood agar plates. All plates were incubated aerobically 24 hours at 37.5°C. before counting the colonies. Each broth containing endotracheal tube was also incubated 24 hours. A second series of plates was prepared, using 0.1 ml. volumes of the incubated broth as a check on the first plates. The numbers of organisms were multiplied by the dilution factor of 1000 to determine the number of organisms per tube. The results of cultures from 12 tubes are shown in Table 16 A (page 89). The following types of organisms were recovered: aerobic spore bearers, Neisseria, *Streptococcus viridans*, coliforms and Micrococci, including *Micrococcus pyogenes var. aureus*.

TABLE 16 A

Degree of Bacterial Contamination of Endotracheal Tubes

Tube Number	Number of Colonies per Tube	
	Before incubation	After incubation
1 (Taken from cupboard)	0	TNTC*
2 " " "	0	TNTC
3 " " "	0	TNTC
4 " " "	0	TNTC
5 " " "	0	TNTC
6 " " "	0	TNTC
7 " " "	0	TNTC
8 " " "	0	TNTC
9 (Sampled after rinsing)	0	0
10 " " "	0	0
11 " " "	0	0
12 " " "	80,000	340,000

* Too numerous to count

Test Series

The first 10 endotracheal tubes in the test series were treated similarly to those in the preliminary series, except that each tube was placed in 10 ml. of Letheen broth and shaken mechanically in a horizontal position for 15 minutes. (The purpose of the shaking was to ensure complete removal of the organisms from the tubes.) 0.1 ml. volumes of broth were then plated on blood agar plates which were incubated aerobically 48 hours at 37.5°C. before counting colonies. The numbers of organisms were multiplied by the dilution factor of 100 to determine the number of organisms per tube. The results are given in Table 16 B (page 91). Two of the cultures (from tubes 15 and 25) yielded Pseudomonas pyocyaneus.

The next 6 endotracheal tubes were cultured in the same way, except that 0.2 ml. volumes of Letheen broth were plated on blood agar plates. The incubation procedure was carried out as before. The results are given in Table 16 B (page 91).

Since relatively small amounts of broth were used to wash a large area and very small amounts of this broth were sampled for growth, the figures can be taken to represent a minimal amount of contamination on the endotracheal tubes and even a small amount of contamination is significant.

TABLE 16 B

Degree of Bacterial Contamination of Endotracheal Tubes

Tube Number	Number of Organisms per Tube	
	Before Incubation	After Incubation
13 (Taken from cupboard)	0	18,000
14 " " "	100	TNTC*
15 " " "	900	TNTC
16 " " "	300	TNTC
25 " " "	TNTC	TNTC
26 " " "	300	TNTC
27 " " "	200	TNTC
28 " " "	TNTC	TNTC
29 " " "	TNTC	0
30 " " "	300	TNTC
19 " " "	100	TNTC
20 " " "	400	TNTC
21 " " "	0	0
22 " " "	50	TNTC
23 " " "	50	TNTC
24 " " "	50	TNTC
No. of Samples	16	
No. TNTC	3	
Average No. of Organisms per Tube	183	

* Too numerous to count

[illegible]

[illegible]

[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
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[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]

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Modified Decontamination Procedure

Since endotracheal tubes were almost always contaminated and the micro-organisms were sometimes those which could cause postoperative infections, it was decided to alter the decontamination procedure. Tubes were thoroughly scrubbed in Germa Medica or pHisohex and soaked overnight in an aqueous Zephiran chloride solution (1/1000). Tubes were rinsed in tap water before use.

Endotracheal tubes and mouthpieces were sampled in the same way as those in the preceding series with 0.1 ml. volumes of 10 ml. Letheen broths plated on blood agar plates after a 15 minute mechanical shaking. Repeat plates were prepared using 0.1 ml. volumes of broth after incubation with the tubes at 37.5°C. for 24 hours. All cultures were incubated aerobically for 48 hours at 37.5°C. before counting the colonies. Cultures of Micrococcus pyogenes var. aureus were isolated for determination of coagulase reactions and phage typing. The results are given in Table 16 C (page 93).

The degree and type of bacterial contamination of tubes receiving the original and modified decontamination treatments are compared in Table 16 D (page 94).

TABLE 16 C

Degree of Bacterial Contamination of Endotracheal Tubes and Mouthpieces

Receiving Modified Decontamination

Sample No.	Number of Organisms per Tube	
Endotracheal Tubes:	Before incubation	After incubation
2 (Sampled from Zephiran soak)	300	0
3 " " "	TNTC*	TNTC
4 " " "	TNTC	TNTC
5 " " "	0	0
7 " " "	100	0
8 " " "	0	0
12 " " "	100	0
13 " " "	0	TNTC
14 " " "	200	TNTC
15 " " "	100	0
16 " " "	200	100
17 " " "	200	0
18 " " "	0	100
Mouthpieces:		
1 (Rinsed with tap water after soaking)	200	TNTC
6 " " "	100	0
9 " " "	0	0
10 " " "	0	100
11 " " "	100	TNTC
19 " " "	300	200
No. of Samples	19	
No. with growth TNTC	2	
Average No. of organisms per tube	100	

* Too numerous to count

TABLE 16 D

Comparison of Degree of Bacterial Contamination of Endotracheal Tubes Before and After Modified Decontamination

	Original Decontamination Procedure	Modified Decontamination Procedure
No. of samples	18	19
No. of samples with *Staphylococcus aureus*	4	1
No. of samples with *Pseudomonas pyocyaneus*	2	1
No. of samples with coliforms	4	0
No. of samples with growth TNTC*	3	2
Average no. of organisms per tube	183	100

* Too numerous to count

Summary of Results

(1) Preliminary cultures of endotracheal tubes showed no growth from 11 of the 12 tubes sampled from the storage cupboard. The culture with growth had a count of 80,000 coliforms per tube.

Nine of the tubes sampled after incubation in broths showed growth. Two of these contained M. pyogenes var. aureus and 2 contained Pseudomonas pyocyaneus.

(2) Sixteen cultures of endotracheal tubes in the test series had an average count of 183 organisms per tube. Three of the cultures had growth too numerous to count. Two cultures contained coliforms, 2 contained Pseudomonas pyocyaneus and 2 contained M. pyogenes var. aureus.

(3) Nineteen endotracheal tubes and mouthpieces decontaminated by the modified procedure had an average count of 100 organisms per tube. Two of the cultures had too many organisms to count. One of the cultures contained Pseudomonas pyocyaneus and one contained coagulase negative M. pyogenes var. aureus. One of the cultures of tubes sampled after incubation contained coagulase positive M. pyogenes var. aureus.

(4) Three cultures of M. pyogenes var. aureus, obtained after the endotracheal tubes were incubated in broth, were phage typed. Each of the following phage types or patterns lysed one culture: 53^+, 3A/6/7/Pool B/C, 3A/6/47/81/Pool C.

INCIDENTAL FACTORS

(j) Blankets

Introduction

Historical

Blankets and blanket dust have been considered a means of spread for pathogenic micro-organisms. Hare and Willets (1941), Willets and Hare (1941), McKissock et al. (1941), Blair (1948) and Hare (1956) have noted the aerial spread of micro-organisms in dust during bed-making. Bourdillon et al. (1948) suggested that all woolen materials should either be oiled or excluded from operating theatres. Clark et al. (1952), Clayton and Robertson (1945), VandenEnde and Walter (1952) and Loosli (1948) recommended oiling for the prevention of blanket dust. Leslie (1953) recommended the use of various types of mineral oil emulsions with or without Fixanol C for controlling the liberation of blanket dust. Harwood et al. (1944) and Barnard (1952) also recommended oil emulsions containing Fixanol C for controlling blanket contamination. Blowers and Wallace (1955) recommended the use of cetyl trimethyl ammonium bromide (C.T.A.B.). Starkey (1956) suggested the use of oil emulsions containing Fixanol C or C.T.A.B., chloramine T or heat treatment for controlling the spread of pathogens by blankets. All workers agreed that blankets are an important source of contamination and that some means of controlling this contamination is required.

Use of Stretcher Blankets in the University of Alberta Hospital

Cotton stretcher blankets and sheets are used for covering all

patients, except infants, in transit to operating theatres. Before entering operating theatres, blankets are folded and placed on the bottom of the stretcher. After surgery the blankets are unfolded and the patient is blanketed in the operating theatre. Stretcher blankets receive no special treatment other than an occasional laundering.

Materials and Methods

Test Series

Blood agar plates were used to culture stretcher blankets by the sweep plate technique (Blowers and Wallace, 1955). Opened culture plates were turned upside down over the folded blankets and rubbed vigorously over an area of approximately 1 square foot. Blankets were sampled immediately after the patient had entered the operating theatre. All cultures were incubated aerobically at 37.5°C. for 48 hours before counting colonies. The results are given in Table 17 A (pages 98, 99). Results are summarized according to the time of sampling and type of contamination in Table 17 B (page 100). Cultures of *M. pyogenes var. aureus* were isolated for determining their coagulase reaction and for phage typing. Since the sampling method does not allow for complete recovery of all organisms present, the results indicate the minimum degree of contamination of stretcher blankets.

TABLE 17 A

Degree of Bacterial Contamination of Stretcher Blankets

Number of Organisms per Square Foot

Date 1956	Case No. 1	2	3	4	5
Mar. 29	313	23			
May 17	223	320		62	82
24	174	115	87	131	
29	232	167	32	160	
30	68	48	218	150	321
June 5	250	167	41	171	
6	242	90	305		
26	132	197	153	145	
27	121	142	122		
July 4	128	81			
5	66	108	36		
25	165	68			
Aug. 8	280	18	156		
9		166	111		
16	84	178			
17	211				
22	63	15	73		
23	122	106	108		93
Sept. 6	36	111	114	36	
7	98	115			

(continued on page 99)

TABLE 17 A (continued)

Date 1956	Case No. 1	2	3	4	5
Sept. 12	50	105		84	
13	155	113		253	
25	51	250			
26	128	198	77		
No. of Samples	23	23	14	9	3
Average No. of Organisms per Square Foot	147	127	117	144	165

TABLE 17 B

Degree and Type of Bacterial Contamination of Stretcher Blankets

Case No.	No. of Samples	No. of Samples Containing *M. pyogenes var. aureus*	No. of Samples Containing *Streptococcus viridans*	No. of Samples Containing *Pseudomonas pyocyaneus*	Average No. of Organisms per Square Foot
1	23	16	10	1	147
2	23	15	10	0	127
3	14	12	7	0	117
4	9	6	6	0	144
5	3	2	3	0	165
Totals	72	51	36	1	
Average no. of organisms per square foot					132

Summary of Results

(1) A total of 72 stretcher blankets were cultured by the sweep plate technique. The average number of organisms per square foot of blanket surface was 132.

(2) Time of sampling had no apparent effect on the degree of contamination of the blankets.

(3) Organisms which grew on the culture plates included Actinomycetes, moulds, aerobic spore bearers, diphtheroids, Gaffkya, yeast, Micrococcus, Pseudomonas and Streptococcus.

(4) *Pseudomonas pyocyaneus* was recovered from one of the blankets.

(5) *Streptococcus viridans* was recovered from 36 of the blankets.

(6) *M. pyogenes var. aureus* was recovered from 51 of the samples. Twenty-two of these cultures were coagulase positive, and 10 were coagulase negative. No coagulase determination was carried out for the remaining 19 cultures.

(7) A total of 38 cultures of *M. pyogenes var. aureus* were phage typed. Fifteen of these were non-typable. Four cultures were lysed by phage type 81. Six were lysed by phage pattern 3A/Pool B and two by 7/53/70/77. Each of the following phage patterns lysed one culture: N.T./Pool C (53), N.T./Pool B (3C), 52A/81/Pool A/C 1/C 2, 47/Pool C1/C2, 52/52A/81, 55/6/7/47/53/54/70/75, 7/47/53/54/42E/75, 7/47/53/54/70/75/77+.

(k) Depots

Introduction

Historical

Depots or fomites have been defined as inanimate objects which are capable of retaining and transmitting infective agents for a long period of time. Gardner et al. (1937) discussed the importance of frequent cleaning of various depots, including walls, overhead lamps, fixtures, furniture and other objects which might act as dust traps. The importance of depots such as contaminated baths was cited in the Medical Research Council War Memorandum (1944). Colbeck (1956) discussed the importance of fomites, especially baths and hand washing facilities. Starkey (1956) stated that such depots as floor joints, ledges, roller blinds, radiators, faucet taps and bar soaps could be eliminated from operating theatres. He suggested frequent washing or oiling of other depots to prevent the spread of infections.

Decontamination of Depots in the University of Alberta Hospital

Cupboards, sinks, lamps, operating tables and other fixtures are washed at the end of each operating day or after infected (dirty) cases, using sponges soaked in aqueous Zephiran chloride solution 1/1000. The lower parts of walls are washed weekly by the nursing staff with a solution containing Germa Medica or Tincture of Green Soap. The upper parts of walls are washed infrequently by the janitorial staff, who are also responsible for cleaning radiators. Inaccessible or hard to reach areas such as high cupboards, X-ray

viewers, radiators or lower parts of furniture may miss cleaning and are sometimes covered with dust. No dust laying methods are used. Floors are cleaned as described in Section (e). Areas of floor beneath immovable furniture frequently escape cleaning.

Materials and Methods

Test Series

Sterile aluminum templates were used in swabbing one inch areas of various depots at the end of the operating day. Swabs were moistened in nutrient broth before swabbing and were immersed in 3 ml. volumes of the broth immediately after. In the laboratory the broths were shaken either manually or mechanically for 15 minutes. 0.1 ml. volumes of the broths were then plated on blood agar plates. All cultures were incubated aerobically at 37.5°C. for 48 hours before counting colonies. Counts were multiplied by the dilution factor of 30 to determine the numbers of organisms per square inch of surface. Cultures of _M_. _pyogenes_ _var_. _aureus_ were isolated for determining their coagulase reactions and phage typing. The results are given in Table 18 A (page 104).

Controls

Various depots were cultured in rooms M 86 and M 71, University of Alberta. These student bacteriology laboratories were used as controls in order that standards for comparing the degree of contamination in operating theatres might be obtained. Swabbing and culturing methods were the same as those described previously. The results are given in Table 18 B (page 105).

TABLE 18 A

Degree of Bacterial Contamination of Depots in Operating Theatres

Location	No. of Samples	No. of samples containing Staph. aureus	No. of samples containing growth TNTC*	Average no. of organisms per square inch of surface
Air conditioner	2	0	0	330
Anaesthesia cabinet	22	3	0	171
Anaesthesia equipment cupboard	25	1	2	105
Base of operating table	21	2	0	1,155
Floor near operating table	35	3	1	1,560
Floor near radiator pipes	14	1	4	1,266
Fly swatter	2	0	0	120
Lamp	29	2	5	471
Sink tops	1	0	0	840
Soap dishes	2	0	0	510
Sponge racks	1	1	0	270
Supply cupboard	13	2	0	480
Walls	5	0	0	1,132
Window sill	19	0	1	354
X-ray viewer	1	1	0	540
Total	192	16	13	
Average no. of organisms per square inch				723

* Too numerous to count

TABLE 18 B

Degree of Bacterial Contamination of Depots in Laboratory

Location	No. of Samples	No. of samples containing Staph. aureus	No. of samples containing growth TNTC*	Average no. of organisms per sq. in. surface
Analytical balance	1	0	0	780
Blackboard ledge	2	0	0	990
Centrifuge lid	1	0	0	900
Cupboard tops	2	1	0	390
Desk	1	0	0	120
Top of first aid cabinet	1	0	0	840
Floor	4	1	0	600
Floor near radiator pipes	2	0	0	4320
Garbage receptacles	3	0	0	920
Incubator top	1	0	0	270
Lab. benches	9	1	0	153
Lab. coat	1	1	0	270
Media cart	1	0	0	60
Radiator	1	0	0	2220
Refrigerator top	1	0	0	840
Shoes	1	1	0	14,520
Sinks	3	2	1	TNTC
Stool	1	0	0	150
Tables	5	2	0	216
Waterbath top	1	0	0	300
Window sills	4	0	0	990
Total	48	9	1	
Average no. of organisms per square inch				943

* Too numerous to count

Summary of Results

(1) 192 cultures were obtained of 1 square inch areas of depots from 15 different locations in operating theatres. Thirteen of the cultures contained growth too numerous to count, while the remaining cultures averaged 723 organisms per square inch.

(2) Depots which were heavily contaminated (over 1000 organisms per square inch) included walls, floors and the bases of operating tables.

(3) Sixteen of the cultures (8%) contained M. pyogenes var. aureus. Three of these were coagulase negative. Streptococcus viridans was contained in 12 of the cultures.

(4) Forty-eight cultures were obtained of 1 square inch areas of depots in the laboratory. One of the cultures contained growth too numerous to count, while the average count of the remaining cultures was 943 organisms per square inch.

(5) Depots which were heavily contaminated (over 1000 organisms per square inch) included floors, radiators, sinks and shoes. Contamination of other depots was of approximately the same degree as that found in operating theatres.

(6) M. pyogenes var. aureus was contained in 9 of the cultures. Three of these were coagulase positive. Streptococcus viridans grew on 4 of the culture plates. Other organisms which grew on the culture plates included aerobic spore bearers, moulds, Actinomycetes, diphtheroids, Neisseria, Gaffkyae and Micrococci.

(7) Eighteen cultures of M. pyogenes var. aureus were phage typed. Nine of these were non-typable. Two of the cultures were lysed by phage type 3A or by phage patterns 29/47/Pool C1/C2 or 47/Pool C1/C2. One of the cultures was lysed by phages 81/52/52A, 7/47/53/54/75/77 or N.T./ Pool B (3A/3C).

(1) Surgeons' Hands

Introduction

Historical

The use of rubber gloves was introduced by Halsted in 1889 for instrument nurses. Surgeons first wore rubber gloves in 1894 (Williams, 1956). Before this period surgeons relied upon mercuric chloride solutions to form an antiseptic barrier on their hands. Adequate preoperative preparation of hands is still important because of the frequency of glove punctures and tears (Blair, 1948; Starkey, 1956). Numerous investigators including Meleney (1935), Beck (1936), Meleney et al. (1940) and Willets and Hare (1941) have listed the surgeon's hands among possible sources of contamination for operative wounds. Pulaski (1947) stated that antisepsis of surgeons' hands and patient's skin are a necessity in surgical procedures.

Innumerable agents have been used for preoperative preparation of surgeons' hands and of the skin of patients since Semmelweis introduced the use of chloride of lime. Mercuric chloride was once used, but was later replaced by potassium mercuric chloride (Price, 1939, 1950). The use of ethyl and isopropyl alcohols of varying percentages has been suggested by Price (1939, 1950), Pulaski (1947) and Squire (1951). Both Price (1950) and Hauser and Cutter (1944) recommended the use of ethyl alcohol to remove soap before soaking hands in Zephiran chloride solutions. Heineman (1937) recommended the use of the quaternary ammonium compound, Zephiran chloride, for preoperative preparation of surgeons' hands. Other quaternary ammonium compounds suggested for the same use were cetyl trimethyl ammonium bromide (Barnes, 1942) and

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Hyamine (Swan et al., 1949). Squire (1951) suggested the use of Dettol for decontaminating hands and Cetrimide for preoperative preparation. Soaps containing hexachlorophene (hexachlorodiphenyl methane) have been recommended by various investigators including Traub et al. (1945), Fuller et al. (1948), Thirlby and Nesbit (1949), Nungester et al. (1949), Freeman and Young (1949, 1950) and Cleland (1952). Blank and Coolidge (1950) stated that the addition of hexachlorophene to soaps was of little value, although Blank et al. (1950) recommended the use of a hexachlorophene scrub combined with a cetyl and isopropyl alcohol soaking. Dull et al. (1950) suggested the use of a Zephiran chloride soak after a hexachlorophene soap scrub. Several of the investigators including Traub et al. (1944), Bowers (1949) and Cleland (1952) stated that shorter scrubs were effective with hexachlorophene soaps, but Price (1951) warned that single short scrubs were inadequate.

Some of the confusion concerning the best agents and scrub techniques resulted from the difficulty in determining the efficiency of skin disinfection measures. Phenol coefficient methods (Ruehle and Brewer, 1931) were said to be adequate for phenol derivatives only (Brewer, 1944). Numerous tests were developed to determine the efficiency of antiseptics in the presence of living tissues. Salle et al. (1937) devised the toxicity index test using chick embryo. Green and Birkeland (1944) and Gershenfeld and Witlin (1947) used modifications of this test. Sarber (1942) and Nungester et al. (1949) used the infection prevention test method. Spaulding and Bondi (1946, 1947) developed an infection prevention toxicity test method. Herrel and Heilman (1943) used a tissue culture method. Skin biopsies have been used by Walter (1938), Helmsworth and Hoxworth (1945), Key (1947), Artz et al. (1951), Murphy

et al. (1951) and Myers et al. (1956). Kraissl (1950) used skin biopsies from guinea pigs. Skin scrapings were used by Robb (1913), Evans et al. (1950), and by Blank and Coolidge (1950). Culture plates taped on treated areas of skin were used by Novak and Hall (1939) and Best et al. (1950). Swabbing was used by Clarke (1942), Barnes (1942) and Hagan et al. (1946) to determine the value of skin antiseptics. Gardner and Seddon (1946, 1948) used organisms added to marked areas of skin, while Story (1952) used glass rings to isolate the test areas of skin surface. Price (1938) devised the serial basin scrub tests to determine the efficiency of agents used in preoperative scrubs. He coined the word "degermation" to describe the killing and removal of micro-organisms from hands. The following investigators have used scrub tests for various skin antiseptics: Cromwell and Leffler (1942), Bernstein (1942, 1948), Hatfield and Lockwood (1943), Traub et al. (1945), Clark et al. (1947), Seastone (1947), Price and Bonnett (1948), Fahlberg et al. (1948), Blank and Coolidge (1950), Harrison and Cockcroft (1952), Canzonetti and Dalley (1952). Hufnagel et al. (1948) and Chisholm et al. (1950) have used mechanical scrubbers to control the pressure intensity of the test scrub procedure.

The various test procedures have added to our knowledge of skin flora, but many problems are still unsolved. Gardner and Seddon (1946) stated that chemical sterilization of skin is impossible without complete destruction of tissue, but Wallace (1949) claimed that sterilization could be accomplished. Price (1938) divided skin flora into transients which were easily removed and residents located in the crypts and crevices of the superficial layers of skin. Lovell (1945, 1946) stated that resident organisms occurred in sebaceous glands and hair follicles

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as well. Resident organisms which are difficult to remove were said to be the "weakest link in the aseptic chain" (Lovell, 1946; Canzonetti and Dalley, 1952). Walter (1952) stated that micro-organisms do not remain on wet skin. Pillsbury (1946, 1952) noted that pathogens may become resident and difficult to remove. Natural factors such as skin fatty acids, sunlight, drying, etc. were said to be important in controlling skin flora (Pillsbury, 1952; Ricketts et al., 1951). Some of the skin antiseptics have been said to act by forming antiseptic barriers under which micro-organisms may multiply (Price, 1939; Blank and Coolidge, 1950). Because of the frequency of glove punctures thorough scrub techniques are necessary regardless of the agent used.

Preoperative Scrubs at the University of Alberta Hospital

Germa Medica (containing 2% hexachlorophene) or PHisohex (pHisoderm with 3% hexachlorophene) are used for all preoperative scrubs. The former is dispensed in containers with foot-control pedals. The latter is provided in plastic squeeze-bottles. The length of the scrub procedure is determined individually and may vary from 1 to 10 minutes depending on the time lapse between scrubs. Nurses follow predetermined scrub routines lasting either 3 or 10 minutes. A typical 3 minute scrub routine follows:

(1) Hands and arms are rinsed with tap water.

(2) Nails are cleaned and hands again rinsed.

(3) Each hand is scrubbed for 1 minute, using a sterile brush and Germa Medica.

(4) Each arm is scrubbed ½ minute.

(5) Hands and arms are rinsed with tap water.

(6) Hands are rinsed in Germa Medica.

(7) Hands and arms are dried using sterile towels.

(8) Gowns are donned, talcum applied to hands and gloves donned.

No effort is made to ensure the exclusive use of hexachlorophene soaps by personnel. Brushes and nail cleaners are provided. These are sterilized and placed in open containers at the beginning of each operating day. Tap water is used for all scrubs. Taps have gauze filters and are controlled by knee-levers.

Materials and Methods

Preliminary Tests

Nine surgeons' hands were swabbed on 2 operating days before scrubbing, after scrubbing and after the case. Swabs moistened in serum broth were used. (Serum broth was employed to inactivate hexachlorophene which might be present on the hands.) Each swab was streaked across the back of the hand 3 times and once down each finger and thumb. The procedure was repeated on the palm of the hand and then on the other hand. Swabs were immersed in 4 ml. volumes of serum broth. In the laboratory 0.1 ml. volumes of broths were plated on blood agar plates after the swabs and broths were shaken either manually or mechanically for 15 minutes. All cultures were incubated aerobically for 48 hours at 37.5°C. before counting colonies. Counts were multiplied by the dilution factor of 40 to determine the number of organisms per swab. The results are given in Table 19 A (page 112).

TABLE 19 A

Degree of Bacterial Contamination of Surgeons' Hands

Date 1956	Subject	Number of Organisms per Swab		
		Before Scrub	After Scrub	After Case
Feb. 23	1 A	2080	80	0
	2 A	2120	0	
	3 A	----	280	240
	3 B	----	520	440
March 1	1 A	4560	0	0
	1 B	40	---	---
	2 A	880	0	0
	3 A	8460	40	0
	3 B	----	80	40
No. of Samples		6	8	7
Average No. of Organisms per Swab		3023	125	103

[illegible]

[illegible]

[illegible]

[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
	[illegible]	[illegible]	[illegible]	
[illegible]	[illegible]	----	[illegible]	
[illegible]	[illegible]	----	[illegible]	
----	----	[illegible]	[illegible]	[illegible]
----	----	[illegible]	[illegible]	
[illegible]	[illegible]	[illegible]	[illegible]	
[illegible]	[illegible]	[illegible]	[illegible]	[illegible]
[illegible]	[illegible]	----	[illegible]	
[illegible]	[illegible]			[illegible]
[illegible]	[illegible]	[illegible]		[illegible]

Test Series

Cultures of surgeons' hands were obtained after scrubbing and after the case. Culturing procedures were the same as those used in the preliminary experiments. Colonies of M. pyogenes were isolated for phage typing and determining coagulase reactions. The results are given in Table 19 B (pages 115, 116, 117 and 118). On 7 occasions surgeons scrubbed twice before operating. The results of both scrubs are given. The results are summarized in Table 19 C (page 119).

Summary of Results

(1) Preliminary cultures of surgeons' hands were obtained by swabbing before scrubbing, after scrubbing and after the case. The average counts were 3032, 125 and 103 micro-organisms respectively. M. pyogenes var. aureus was contained in one of the samples.

(2) 196 cultures of surgeons' hands were obtained after pre-operative scrubs. Fourteen of the cultures contained too many organisms to count. The remaining cultures contained an average of 974 micro-organisms per swab. M. pyogenes var. aureus was contained in 39 of the samples. Coagulase tests were done on 19 of the cultures. Only 3 cultures were coagulase positive.

(3) Seven cultures of surgeons' hands were obtained after a second preoperative scrub. Three of the cultures contained too many organisms to count. The average count of the remaining 4 was 225 micro-organisms per swab. Three of the cultures contained M. pyogenes var. aureus.

(4) 122 cultures of surgeons' hands were obtained after the operative procedure. Seven of these had too many organisms to count,

while the remaining 115 had an average of 636 micro-organisms per swab. M. pyogenes var. aureus was contained in 12 cultures. Coagulase reactions were determined for 6 of the cultures. Three were coagulase positive.

(5) Preoperative scrubs resulted in an approximate reduction of 96%. The number of organisms was lower after the case than after the scrub, contrary to results reported by other investigators. This was probably due to the action of hexachlorophene which forms a protective film on the hands.

(6) Yeast was recovered from one culture obtained after scrubbing and two cultures obtained after the case. Other organisms which grew on the culture plates included diphtheroids, aerobic spore bearers, Neisseria, Gaffkya, Micrococcus and Actinomycetes.

(7) Thirty-three cultures of M. pyogenes var. aureus isolated from surgeons' hands before scrubbing were phage typed. Of these twenty-four were non-typable. One culture was not lysed by individual bacteriophage strains, but was lysed by Pool A. Each of the following phage types or patterns lysed one culture: 3A, 29/73, 29/47/Pool C1/C2, 7/53/70, 6/7/47/53/73/81, 6/7/47/53/81, 52/52A/Pool A, 47/Pool C1/C2.

Seven cultures of M. pyogenes var. aureus isolated from surgeons' hands after the case were phage typed. Five were non-typable and of the remaining 2 cultures one was lysed by phage pattern 29/47/Pool C1/C2 and one by phage pattern 47/Pool C1/C2.

In all four samples in which cultures obtained both after scrubbing and after the case were phage typed both cultures were lysed by identical phage types or patterns.

TABLE 19 B

Degree of Bacterial Contamination of Surgeons' Hands

Number of Organisms per Swab

Date 1956	Subject	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
		After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case
Feb. 23	A	80	0	0		280	240				
	B					520	440				
Mar. 1	A	0	0	0	0	40	0				
	B					80	40				
Mar. 8	A	0		200	0	0					
	B	80	15760	40	40	280	0				
Mar. 15	A	80	800	40	40	40	40				
	B				40	320	80				
Mar. 22	A	80	160	0	160						
	B	80/TNTC*	TNTC	680							
	C			TNTC/TNTC							
Mar. 29	A	80	TNTC	TNTC	0	TNTC					
	B	4400/TNTC		1520		120	280				
	C	TNTC				TNTC					
May 17	A	600	80	0		880	480	2240	2440	160	240
	B			0	40						
May 24	A	40		120		TNTC	0	560	7880		
	B	TNTC	TNTC	80	TNTC	5200					

* Too numerous to count

(continued on page 116)

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[illegible]

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TABLE 19 B (continued)

Date 1956	Subject	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
		After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case
May 29	A	40	40	1080		40		3120	320	600	160
	B	TNTC	80							40	80
	C	240	40							0	0
May 30	A	1960		160	80	1720	1200			520	4600
	B	320	40	1080	80	0	240				
June 5	A	0	40	80		80	80	160	440		
	B	1640	1400	2440		40		160			
	C	18,240		240	480	TNTC	0				
June 6	A	1560				1280	560				
	B	0				160					
	C					0	40				
June 26	A	40	40	40	0	240		360			
	B	TNTC	320			TNTC		1200			
	C		40			40			120		
June 27	A	40	0	0		80	40	120	120		
	B	0		400	80	0					
	C	0		120	40						
July 4	A	0	40								
	B	960									
July 5	A	200/160		1080		160					
	B	920	1200	200		0	0				
	C						TNTC				
	D					12920					

(continued on page 117)

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TABLE 19 B (continued)

Date 1956	Subject	Firth Case		Second Case		Third Case		Fourth Case		Fifth Case	
		After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case
July 25	A	4200	280	520	0						
	B	200	0	1480	80						
	C	200		400							
	D	1400	0	40	0						
	E	4600									
Aug. 8	A	400		0	80	320	80				
	B	280	160	600		80	520				
	C	800	200	3560	160	680	600				
Aug. 9	A	9600	1160	12800	0	800					
	B	240	120	80	40	1880	160				
	C			40		16000					
	D			520	440						
Aug. 16	A	280	80								
	B	2720									
Aug. 17	A	480	0								
	B	200	40								
Aug. 22	A	320	80	560		2080	80				
	B		3800	80		40	200				
Aug. 23	A		160	80	40	1800		440	1160	280	40
	B	960								840	
Sept. 6	A	960	160	80		120		TNTC			
	B			240	720			200	280		
	C			600							

(continued on page 118)

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TABLE 19 B (continued)

Date 1956	Subject	First Case		Second Case		Third Case		Fourth Case		Fifth Case	
		After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case	After Scrub	After Case
Sept. 7	A	6640		480		600	40	240	80	TNTC	40
	B					1360	120	40	40	840	
	C							160		120	TNTC
Sept. 12	A	80/0		0	80	40/560	TNTC	1960	160		
	B	1520		80	18,240	3120					
	C	40				200	160				
Sept. 13	A	240/80		1000	560	320	0				
	B	520	160	TNTC		1240					
	C	40	80	80		120					
Sept. 25	A	3000	200	40							
	B	40	120	0	0						
	C	120		160							
Sept. 26	A	840	160	200	120	120					
	B	0	120	0	280	0	0				
	C	160	0	560	120	870					
No. of Samples		67	42	56	32	55	29	15	11	10	8
No. with Growth TNTC		6	3	4	1	5	2	1	0	1	1
Average No. of Organisms per Swab		1197	694	652	711	1137	212	783	1185	378	737

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TABLE 19 C

Comparison of Type and Degree of Bacterial Contamination of Surgeons' Hands

	Before Scrub	After First Scrub	After Second Scrub	After Case
No. of Samples	6	196	7	122
No. with growth TNTC*	0	14	3	7
No. containing *M. pyogenes var. aureus*	1	39	3	12
Average no. of organisms per swab	3023	974	225	636

* Too numerous to count

(m) Patients' Skin

Introduction

Historical

The skin of patients has been recognized as a source of pathogenic micro-organisms since the introduction of antiseptic techniques in surgery (Trent, 1946; Williams, 1956). Lovell (1946) and Canzonetti and Dalley (1952) stated that the preoperative preparation of skin is still one of the weakest links in the aseptic chain. Meleney (1935), Meleney et al. (1940) and Willets and Hare (1941) discussed the importance of skin contamination by hemolytic streptococcal carriers. Miles et al. (1944) stated that 10 to 20% of patients were skin carriers of _M. pyogenes var. aureus_. Williams and Miles (1949) stated that up to 50% of the population were skin carriers of _M. pyogenes var. aureus_ and approximately 4% were carriers of _Streptococcus pyogenes_. Barber et al. (1949) found skin carrier rates for _M. pyogenes var. aureus_ varying from 10 to 51%. Blair (1948) stated that the skin of the body is less heavily contaminated than exposed areas, but still requires adequate preparation. Hare (1956) and Starkey (1956) discussed the importance of preoperative skin preparation.

The first agent used for preoperative skin preparation was phenol. Later mercuric chloride and alcohol-ether dressings were used (Brewer, 1915). Iodine, one of the earliest agents (Brewer, 1915) was used frequently (Beck, 1936; Allen, 1929) and has received renewed interest (Gardner, 1948; Garrod, 1948; Price, 1950). Ethyl alcohol has been used in iodine preparations. Both ethyl and isopropyl alcohols are often used alone in varying percentages or in combination with other drugs

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(Pulaski, 1947; Archer, 1945; Straughn, 1946; Price, 1950). Flavine dyes were introduced during the Second World War (Bonny and Allen, 1944). Quaternary ammonium compounds including benzalkonium chloride ("Zephiran chloride") (Wright and Wilkinson, 1939; White et al., 1938) cetyltrimethyl ammonium bromide (Barnes, 1942), cetylpyridinium chloride or "Ceepryn" (Clarke, 1942; Kramer and Sedwitz, 1944; Brown et al., 1944) have been used for preoperative skin preparation. Guild (1945) suggested the use of pHisoderm. Numerous workers have recommended the use of soaps containing hexachlorophene, but others (Dull et al., 1950; Fahlberg et al., 1948) warn that serum or plasma inactivates the agent. Hibitane is one of the newer agents investigated for preoperative skin preparation (Myers et al., 1956).

Preoperative Skin Preparation at the University of Alberta Hospital

The operative site is washed and shaved, if necessary, the night before the operative procedure.

For routine cases the area is swabbed with ether by a "non-sterile" nurse using sterile gauze sponges and forceps. Towels are then placed around the area which is painted with Tincture of Zephiran Chloride 1/1000 by a "sterile" nurse or surgeon using gauze sponges and forceps. Care is taken that all of the operative site is treated. The operative site is draped immediately and the first incision is made. The time lapse between the skin treatment and the incision varies from 1 to 10 minutes depending on the length of time required for draping. No effort is made to control this time lapse. The same skin treatment is used before the administration of spinal anaesthetics.

Germicidal Detergent (Parke-Davis) containing Phemerol was once used

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for orthopedic cases, but is now replaced with an alcohol-iodine treatment. The operative site is swabbed with 50% ethyl alcohol. Towels are then applied and the area is swabbed with 2% Tincture of Iodine in 50% alcohol. After the operation the iodine is removed using 50% alcohol.

Germa Medica is used for obstetrical cases. The agent is swabbed on full strength before draping.

Materials and Methods

Preliminary Tests

Areas of skin approximately 1 square inch in size were swabbed before skin preparation, after treatment and after the case. Swabs were immediately immersed in 4 ml. volumes of broth contained in screw-capped vials. Letheen broth was used for swabs of skin prepared with Tincture of Zephiran. Sodium thiosulphate broth was used for the Tincture of Iodine preparation (see Appendix B). The ends of the swabs were clipped off using sterile scissors and the vials were sealed. In the laboratory the broths were shaken either mechanically or manually for 15 minutes. 0.1 ml. portions of broths were then plated on blood agar plates. All cultures were incubated aerobically for 48 hours at 37.5°C. before counting colonies. Counts were multiplied by the dilution factor of 40 to determine the number of organisms per square inch of skin surface. The results are given in Table 20 A (page 123). No further tests were done.

TABLE 20 A

Degree of Bacterial Contamination of the Operative Field

Date 1956	Case No.	Skin Preparation	Number of Organisms per Square Inch of Skin Surface		
			Before Treatment	After Treatment	After Case
Mar. 1	1	Tr. Iodine	40	0	80
	2	Tr. Iodine	160	0	0
	3	Tr. Iodine	4920	4960	120
Mar. 8	1	Tr. Iodine		0	0
	2	Tr. Iodine		40	80
	3	Tr. Zephiran		680	1400
No. of Samples			3	6	6
Average no. of organisms per square inch of skin surface			1707	947	280

Summary of Results

(1) Six preliminary cultures of 1 square inch areas of skin prepared with Tincture of Iodine or Tincture of Zephiran showed average counts of 1707 micro-organisms before treatment, 947 after treatment and 280 after the case.

(2) Three cultures taken after treatment and two taken after the case showed no growth.

(3) Organisms which grew on the culture plates included aerobic spore bearers, Actinomycetes, diphtheroids, Neisseria, Micrococcus and *Streptococcus viridans*.

(n) Respiratory Tract Flora of Personnel

Introduction

Historical

The apparent increase in nasopharyngeal carriers of antibiotic-resistant staphylococci and their relation to epidemics of staphylococcal infections have been of interest to numerous investigators. Techniques used in studying respiratory flora have included antibiotic sensitivity (determined by sensitivity discs, tube dilution or plate screening methods), serological and bacteriophage typing. The majority of investigators used both antibiotic sensitivity and phage typing in identifying strains of *M. pyogenes var. aureus*. Varying percentage carrier rates for *M. pyogenes var. aureus* have been recorded, from 40% (Gould and Allan, 1954), 55% (Spink, 1954), 60% (Colbeck, 1949) to 76% (Lepper et al., 1955). Carrier rates of up to 90% have been recorded for infants in maternity wards. Nasal carrier rates of approximately 50% (Miles et al., 1944; Williams and Miles, 1949) have been recorded for non-hospital populations. Knight and Collins (1955) and Brodie et al. (1956) have indicated a tendency toward increased carrier rates among hospital personnel and a tendency for such persons to acquire antibiotic-resistant strains of staphylococci which predominate in the hospital environment. Colbeck (1949), Finland and Haight (1953), Sherman et al. (1956), Tulloch (1954), Gould and Allan (1954) and Forfar (1955) have related infection outbreaks to increased carrier rates. Landy et al. (1954) have traced an outbreak of postoperative infections to a single carrier. Treatment of normal carriers of staphylococci has been recommended by Finland and Haight (1953), Gould

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and Allan (1954), Tulloch (1954) and Lowbury (1955). Isolation of infected patients and routine investigation of personnel and patients' respiratory flora as a means of preventing epidemics of staphylococcal infections have been recommended by McGuiness and Musgrove (1949) and by Sherman et al. (1956). The importance of adequate investigation and effective treatment of infections have been stressed by the Southern Medical Journal (1953) and The Lancet (Leading Article, 1955, 1956). Duff and Murray (1953) have warned of increased clinical and bacteriological problems resulting from unrestricted use of antibiotics. Lepper et al. (1953) also suggested that the use of antibiotics should be controlled. Howe (1954) stated that many factors are involved in staphylococcal epidemics, including increased antibiotic resistance and carrier rates. He suggested that increased care in masking and gloving could prevent infections. Clough (1955) stated that staphylococcal epidemics could be controlled by rigid asepsis, dust control measures and adequate, effective treatment of patients. McDermott (1956) noted high carrier rates of staphylococci (50 to 80%), but stated that infection rates were not influenced by normal carriers. He believes that modern therapy is responsible for reducing patients' resistance to infections. The preponderance of the organisms involved and a shift in the delicate host-parasite relationship were also listed as influencing factors.

<u>Investigation and Treatment of Carriers at the University of Alberta Hospital</u>

Respiratory tracts of operating theatre personnel were at one time routinely swabbed before admission to the operating or maternity wings. Carriers of *M. pyogenes* var. *aureus* were treated by oral and topical administration of

chloramphenicol. This procedure is still carried out in the maternity wing, although regulations have been relaxed in the operating wing. Streptococcal carriers (Lancefield Groups A and C) are excluded from the operating theatres until free of the organisms. Carriers go undetected since investigation is spasmodic and infrequent.

Materials and Methods

Noses and throats of personnel were cultured with swabs moistened in serum broth. In the laboratory all swabs were streaked on blood agar plates, ½ plate being used to culture organisms from each subject's nose and the other ½ used to culture organisms from the same individual's throat. All plates were incubated aerobically for 48 hours at 37.5°C. before being examined. Colonies of *M. pyogenes var. aureus* were isolated for determining antibiotic sensitivity, coagulase reactions and phage typing (only coagulase positive strains were phage typed). Antibiotic sensitivity was determined using the disc method with micro-organisms grown on nutrient agar plates. The antibiotics used were Penicillin (P), Erythromycin (E), Chloramphenicol (C), Tetracyclines (T) and Streptomycin (S). Coagulase reactions were determined using a loopful of culture from a nutrient agar plate or 0.5 ml. of a 24 hour nutrient broth culture with 0.5 ml. sterile human plasma. The mixture was examined after 1 and 3 hours incubation for clotting. Results are given in Table 21 A (pages 128, 129) (Nurses); Table 21 B (pages 130, 131) (Surgeons); Table 21 C (page 132) (Internes); Table 21 D (pages 133, 134) (Other Personnel). The results are summarized in Table 21 E (pages 135, 136).

TABLE 21 A

Incidence of M. pyogenes var. aureus Carriers Among Operating Theatre Personnel

(Nurses)

Subject	Presence of S. aureus (pos. or neg.)	Site of Carriage	Coagulase Reaction (pos. or neg.)	Antibiotic Sensitivity (PECTS)	Phage Typing	Other Organisms
A. Nursing Supervisors						
15	neg.					
16	pos.	nose	pos.	ECTS	3A	
17	neg.					coliforms (nose)
41	neg.					
45	neg.					
B. Graduate Nurses						
33	pos.	nose	pos.	PECTS	55	
44	neg.					
7	neg.					
8	neg.					
14	neg.					coliforms (nose)
30	pos.	nose	neg.	PECTS		
31	neg.					
51	neg.					
C. Junior Graduate Nurses						
56	neg.					coliforms (nose)
43	neg.					
44	pos.	nose & throat	pos.	ECTS	N.T.	coliforms (nose)
46	pos.	nose	neg.	PECTS		

(continued on page 129)

TABLE 21 A (continued)

Subject	Presence of S. aureus (pos. or neg.)	Site of Carriage	Coagulase Reaction (pos.or neg.)	Antibiotic Sensitivity (PECTS)	Phage Typing	Other Organisms
D. Student Nurses (Second Year)						
1	pos.	nose	pos.	PECTS	29/52/7+	
2	neg.					coliforms (nose)
5	neg.					
6	neg.					
9	pos.	nose	neg.	PECTS		
10	pos.	nose	neg.	PECTS		
11	neg.					
12	neg.					coliforms (nose)
13	pos.	nose	pos.	PECTS	3A	
19	pos.	nose	pos.	ECTS	7/47/54/70/73/75	
32	pos.	nose	pos.	ECTS	N.T.	
33	pos.	nose	pos.	PECTS	3A/3B/3C/55	
34	neg.					
35	neg.					
36	neg.					
37	pos.	nose	neg.	EC		coliforms (nose)
38	neg.					
39	neg.					
40	pos.	nose & throat	pos.	ECTS	3A	
42	neg.					
47	pos.	throat	pos.	ECTS	7/47/53/54/70/75/77+	

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TABLE 21 B

Incidence of M. pyogenes var. aureus Carriers Among Operating Theatre Personnel

(Surgeons)

Subject	Presence of S. aureus (pos. or neg.)	Site of Carriage	Coagulase Reaction (pos. or neg.)	Antibiotic Sensitivity (PECTS)	Phage Typing	Other Organisms
A. Staff Surgeons						
20	pos.	nose	neg.	ECTS		
23	pos.	nose & throat	pos.	PECTS	N.T.	
25	pos.	nose & throat	pos.	PECTS	7/53	
50	neg.					
84	pos.	nose	neg.	ECTS		
89	neg.					
91	pos.	nose	pos.	PECTS	81/52/52A	
98	pos.	nose	pos.	PECTS	52/52A/6/47/53/54/75+	
99	pos.	nose	pos.	PECTS	29/52/52A/79/81+	coliforms (throat)
101	neg.					coliforms (nose)
B. Resident Surgeons						
22	pos.	nose	pos.	ECTS	81	
73	neg.					

(continued on page 131)

TABLE 21 B (continued)

Subject	Presence of S. aureus (pos. or neg.)	Site of Carriage	Coagulase Reaction (pos. or neg.)	Antibiotic Sensitivity (PECTS)	Phage Typing	Other Organisms
C. Surgeons						
64	pos.	nose	pos.	ECTS	N.T.	
71	neg.					
72	neg.					
74	neg.					
79	neg.					
80	neg.					
81	pos.	nose	neg.	ECS		yeast (throat)
86	neg.					
92	pos.	nose	pos.	PECTS	7/47/53/54/42E/75/77+	
97	neg.					
100	neg.					
110	pos.	nose	pos.	PECTS	N.T.	
118	neg.					
112	neg.					
113	neg.					
114	pos.	nose	pos.	ECTS	6/47/53	
116	pos.	nose	pos.	ECT	7/53/54/70	
D. Medicine						
75	neg.					
106	pos.	nose	pos.	ECTS	47/54+	

TABLE 21 C

Incidence of M. pyogenes var. aureus Carriers Among Operating Theatre Personnel

(Internes)

Subject	Presence of S. aureus (pos. or neg.)	Site of Carriage	Coagulase Reaction (pos. or neg.)	Antibiotic Sensitivity (PECTS)	Phage Typing	Other Organisms
Senior Assistant Resident						
90	pos.	nose	pos.	EC	29	
Assistant Residents						
88	pos.	nose	pos.	ECTS	29/73+	
117	neg.					
Junior Assistant Residents						
21	neg.					
26	pos.	nose	pos.	PECTS	---	
29	neg.					coliforms (nose)
82	pos.	nose	pos.	ECTS	3A/3C	
94	neg.					
107	pos.	nose	neg.	PECS	N.T.	
108	pos.	nose & throat	pos.	ECTS	3A/3B/3C	
109	neg.					
111	neg.					
115	neg.					
Internes (First Year)						
66	neg.					
67	neg.					coliforms (throat)
83	neg.					
85	neg.					coliforms (nose)
87	neg.					
93	neg.					
96	pos.	nose	pos.	PECTS	29/52/52A/73	

TABLE 21 D

Incidence of M. pyogenes var. aureus Carriers Among Operating Theatre Personnel and Students

(Other Personnel and Students)

Subject	Presence of S. aureus (pos. or neg.)	Site of Carriage	Coagulase Reaction (pos. or neg.)	Antibiotic Sensitivity (PECTS)	Phage Typing	Other Organisms
Anaesthetists						
18	pos.	nose	neg.	PECTS		*Streptococcus pyogenes*, Group A (throat)
24	pos.	nose	pos.	PECTS	6/7/47/53/54/42E/70/75+	
27	neg.					
28	neg.					
61	neg.					
62	pos.	nose	pos.	EC	---	
63	neg.					
95	pos.	nose	neg.	ECTS		
103	neg.					
Orderlies						
48	neg.					
49	pos.	nose	pos.	ECS	81/52/52A	
65	pos.	nose	pos.	PECTS	N.T.	

(continued on page 134)

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TABLE 21 D (continued)

Subject	Presence of S. aureus (pos. or neg.)	Site of Carriage	Coagulase Reaction (pos. or neg.)	Antibiotic Sensitivity (PECTS)	Phage Typing	Other Organisms
Secretaries						
57	pos.	nose	pos.	PECTS	6/47/53	
58	pos.	nose	pos.	PECTS	N.T.	
59	pos.	nose	neg.	PECTS		
Students (Medical and Dental)						
69	neg.					
70	neg.					
76	pos.	nose	pos.	PECTS	29/52	
77	neg.					
78	pos.	nose	pos.	PECTS	29/52/79/6/7/47/53/54/42E/70/75+	
104	neg.					
105	pos.	nose	pos.	ECTS	7/42E/70/73	

TABLE 21 E

Incidence of Carriers and Types of M. pyogenes var. aureus Among Operating Theatre Personnel

Position	No. of Subjects	Coagulase Positive M. pyogenes var. aureus		Coagulase Negative M. pyogenes var. aureus		Total Carriers	
		No.	Percent	No.	Percent	No.	Percent
Nurses							
Nursing Supervisors	5	1	20.0	0		1	20.0
Graduate Nurses	8	1	12.5	1	12.5	2	25.0
Junior Graduate Nurses	4	1	25.0	1	25.0	2	50.0
Student Nurses	21	7	33.3	3	13.3	10	47.6
	38	10	26.3	5	13.2	15	39.5
Surgeons							
Staff Surgeons	10	5	50.0	2	20.0	7	70.0
Resident Surgeons	2	1	50.0	0		1	50.0
Surgeons	17	5	29.4	1	5.9	6	35.3
Medicine	2	1	50.0	0		1	50.0
	31	12	38.7	3	9.7	15	48.4

(continued on page 136)

[illegible]

TABLE 21 E (continued)

Position	No. of Subjects	Coagulase Positive M. pyogenes var. aureus		Coagulase Negative M. pyogenes var. aureus		Total Carriers	
		No.	Percent	No.	Percent	No.	Percent
Internes							
Senior Assistant Resident	1	1	100.0	0		1	100.0
Assistant Resident	2	1	50.0	0		1	50.0
Junior Assistant Residents	10	3	30.0	1	10.0	4	40.0
Internes (First Year)	7	1	12.5	0		1	12.5
	20	6	28.5	1	4.8	8	33.3
Others							
Anaesthetists	9	2	22.2	2	22.2	4	44.4
Orderlies	3	2	66.7	0		2	66.7
Secretaries	3	2	66.7	1	33.3	3	100.0
Students	7	3	42.9	0		3	42.9
[illegible]	22	9	40.9	3	13.6	12	54.5
Total Number of Subjects	111	37		12		49	
Percentage of Carriers			33.3		10.7		44.0

Summary of Results

(1) A total of 111 cultures of noses and throats of operating theatre personnel were obtained.

(2) Ten of 38 nurses (26%) were carriers of coagulase positive *M. pyogenes var. aureus*. Five nurses (13%) were carriers of coagulase negative strains.

(3) Twelve of 31 surgeons (38%) were carriers of coagulase positive *M. pyogenes var. aureus*. Three (9%) carried coagulase negative strains.

(4) Six of 20 internes (28%) were carriers of coagulase positive *M. pyogenes var. aureus*. Only 1 (5%) carried a coagulase negative strain.

(5) Nine of 22 other personnel and students (41%) were carriers of coagulase positive strains of *M. pyogenes var. aureus*. Three (14%) were carriers of coagulase negative strains.

(6) A total of 37 (33%) of the 111 subjects were carriers of coagulase positive strains of *M. pyogenes var. aureus* and 12 (11%) were carriers of coagulase negative strains.

(7) The influence of duration of service was best shown in the interne and surgeon groups whose carrier rates increased with their status.

(8) *Streptococcus pyogenes* (Group A) was isolated only once from the throat of an anaesthetist. Micro-organisms of the coliform group were isolated from nose cultures of 10 subjects and throat cultures of 3 subjects. Yeast was isolated from the throat culture of one surgeon.

DISCUSSION

DISCUSSION

A number of difficulties were encountered during the course of this investigation. One of these was the low degree of contamination expected for most of the factors investigated. Sampling was made more difficult because both qualitative and quantitative determinations were attempted. Swabbing was used in many of the culturing procedures. It is recognized that sampling by this method is not very efficient, therefore our results indicate minimum amounts of contamination present. Another problem was the difficulty in standardizing culture methods and conditions under which samples were obtained. A number of variables could not be controlled. The purpose of the investigation, however, was an evaluation of techniques as practised under normal working conditions rather than a series of carefully controlled artificial circumstances.

Average air counts were approximately double the arbitrary limits set by Bourdillon et al. (1948) for major operations. The degree of aerial contamination may actually have been much higher, since neither the air-sampler nor exposed culture plates allow for the breaking up of bacterial clumps. Although it may be argued that a relationship between the degree of aerial contamination and infection rates would be difficult to prove, a greater number of organisms present would indicate a greater possibility for wound contamination. Measures aimed at controlling aerial contamination would be hampered by conditions which cannot be easily improved, including shortages of operating theatres, nursing personnel and equipment. The construction of the operating wing which has no teaching theatres, no separate scrub rooms and no buffer zones between the corridor and operating theatres

also limits the efficiency of any control measures introduced. Despite these drawbacks many improvements could be made in techniques which would help to reduce aerial contamination. These are listed below: --

(1) Separation of clean and "dirty" (infected) cases with the use of one operating theatre for the latter when possible.

(2) More time allowed between operations to allow for "settling out" of particulate matter disturbed during cleaning.

(3) All necessary equipment should be on hand before the operation commences.

(4) Control of traffic and activity in operating theatres.

(5) Operating theatre doors should be closed to reduce contamination from corridors.

Certain control measures already in use might be altered for greater efficiency. For example, air conditioning units could be operated for the intake of outside air rather than recirculation of inside air since the former has been shown to contain fewer pathogens. If overboots are to be worn they should be worn by all personnel and should be donned outside operating theatres rather than in the surgeons' dressing room.

Adequate masking is of extreme importance because of the number of carriers of _M. pyogenes var. aureus_ and occasional carriers of _Streptococcus pyogenes_. All persons entering operating theatres should be properly masked. Masks should cover the nose. Masks should be tightly secured in place and should be changed frequently. Masks should be worn during cleaning of operating theatres because the organisms emitted may still be suspended during the next case or may remain in the dust of the room for a prolonged period of time.

Splash basins are a factor that has usually been overlooked as a

source of contamination. Micro-organisms from this source are of significance because the water comes into direct contact with the operative wound via hands, instruments and sponges. Contamination from this source could be controlled simply by avoiding exposure of basins until they are actually required for use and by changing the water frequently, probably at half hour intervals.

Sheets used on operating tables are probably not of direct importance since they are changed frequently, but they are possibly an indirect source of contamination. Care should be taken in changing sheets and dust control measures using Fixanol C in oil emulsions or some similar agent would help reduce aerial contamination. Because micro-organisms can readily pass through moistened materials more care should be taken in decontaminating operating table mats, which may frequently be contaminated through sheets soaked with blood or exudates.

The very high degree of contamination found in mops, scrub water and floors indicates the need for revision of cleaning techniques. The progressive decrease in contamination during the operating day shown in the results suggests that the source of contamination was probably the mops. Daily washing and daily sterilization would reduce contamination of mops. The substitution of Germa-Medica with another agent (for example, one of the cresol type or Fixanol C), which is not inactivated by blood or serum and is capable of dealing with the degree of contamination encountered would help in controlling contamination. More frequent changing of scrub water would also be of value.

It has been shown that contamination of anaesthesia equipment is important. Prompt treatment is especially desirable. The present method of decontamination (Germa-Medica scrub with a Zephiran chloride soak) is effective if carried out properly. Masks occasionally receive

only a momentary dip in the Zephiran solution. Weekly treatment of connecting tubes and gas machines would also be desirable. Aseptic techniques are usually not considered in anaesthesia. There is no reason why aseptic principles should not be part of the anaesthetist's regimen.

Stretcher blankets are almost always contaminated with pathogens, therefore dust control treatment with Fixanol C oil emulsions and weekly sterilization using hot air ovens are absolutely necessary. Blanketing should be done in halls to prevent the spread of dust in operating theatres.

Depots which could be eliminated from operating theatres include roller blinds and fly swatters. Insecticidal aerosols have been recommended to replace the latter. More frequent, controlled decontamination of depots which cannot be eliminated is important. Hard to clean areas such as radiators, floors below cupboards, cupboard tops, etc. should be more carefully treated.

Cultures of surgeons' hands showed that preoperative scrubs were usually effective, but occasionally a large number of micro-organisms was recovered after the preoperative scrubs. The active agent used (hexachlorophene) is effective if used exclusively, but thorough scrubbing is still required. Decontamination of hands after handling infected materials is still as important as it was in Semmelweis' day.

Cultures of patients' skin were too few for absolute evaluations, but they indicate that the skin was not always sterile. Time is also an important factor. It is important for the surgeon to realize that no agent can act instantaneously. The minute gained is not worth risking infection. Agents used must be chosen carefully. Germa-Medica is not the best agent for obstetrics, since it is readily inactivated

by organic matter.

Routine culturing of respiratory tracts of personnel should be carried out more frequently. Carriers of *M. pyogenes var. aureus* should be treated or warned to be more careful in their masking. Streptococcal carriers are fewer in number, but they are also of importance and can be easily treated.

One of the difficulties in establishing new routines appears to be the lack of a single person or group of persons responsible for co-ordinating the work of the many departments involved, who could enforce and control any modifications in technique.

The results of our investigation agree with the conclusions arrived at by such investigators as Starkey (1956) and McDermott (1956), who point out the ubiquitous character of the organisms currently involved in hospital infections. Starkey states that no-~~one~~ single control measure is adequate but all measures must be followed concurrently. Pathogenic organisms were isolated from cultures of most factors studied. The actual demonstration of pathogens is not essential, but the degree of contamination indicates the relative possibility of wound contamination. Application of aseptic and antiseptic principles is the responsibility not only of the hospital administration, but of every practising surgeon.

Significance of Phage Typing

The results of bacteriophage typing of cultures of _M. pyogenes var. aureus_ show a fairly even distribution of phage types and patterns for approximately one-half of the cultures tested. The remaining cultures did not show lysis with the bacteriophage strains used. Because of the wide variety of types and the large number of factors investigated it would be impossible to incriminate a single factor or strain of _M. pyogenes var. aureus_ as being responsible for any particular hospital infection.

Provincial Laboratory records were available for the first fifty operations investigated at the time the investigation was concluded. These were kindly made available by Dr. R.D. Stuart, Director of the Provincial Laboratory of Public Health and Head of the Department of Bacteriology. Cultures were submitted for only two infections, both of a serious nature involving thoracic cases. The first contained _Clostridium welchii_, _Pseudomonas pyocyaneus_ and _M. pyogenes var. aureus_. The latter was sensitive to penicillin, erythromycin, chloramphenicol, tetracyclines and streptomycin by the disc test method. The second culture contained _M. pyogenes var. aureus_ which was sensitive to erythromycin and chloramphenicol. Unfortunately the organisms from both cultures were non-typable. Non-typable strains of _M. pyogenes var. aureus_ were recovered from the air of the operating theatre before, during and after both cases. They were also recovered from stretcher blankets, splash basins, and from a supply cupboard during the second case. Other typable strains of _M. pyogenes var. aureus_ were also recovered from the air, floors, the surgeon's hands and the

[illegible]

[illegible]

[illegible]

[illegible]

operating table sheet during this case. Because the organisms were non-typable, it would be impossible to definitely associate organisms isolated in the operating theatre with those isolated from the infective processes. The fact that both infections were serious suggests that cultures were possibly not submitted for other more trivial infections.

At one time a large proportion of strains of M. pyogenes var. aureus submitted by the University of Alberta Hospital were phage type 81. The latest records (March 1957) show that type 81 represented one third of the isolations from local sources other than the University of Alberta Hospital, but that cultures from the University of Alberta Hospital were divided among a number of phage types and patterns.

SUMMARY

SUMMARY

Aseptic and antiseptic techniques as practised in the operating theatres of the University of Alberta Hospital were investigated under normal working conditions in the absence of any indication of an outbreak of infection.

General environmental conditions were investigated by air sampling and by determining the effects of traffic, air conditioning, temperature and relatively humidity on aerial contamination. Samples were obtained by use of the G.E. Electrostatic Bacterial Air-Sampler and by exposure of blood agar plates. Average counts during 105 cases representing 35 operating days were 0.9 organisms per cubic foot in the unoccupied theatres, 19.5 before the first case, 13.8 to 16.8 during cases, 19.9 to 21.2 between cases and 15.6 after the last case. The number of organisms settling on exposed culture plates was 8.9, 66.4, 39.8 to 46.4, 40.9 to 46.4 and 273 per hour for the same sampling times. _M. pyogenes var. aureus_ was recovered from two-thirds of the air samples. Air conditioning units caused a gradual increase in aerial contamination during the operating day, especially during cleaning of the operating theatres, probably because particulate matter disturbed at this time was prevented from "settling out". The effect of traffic was inconclusive, but there was a suggestion that increased traffic resulted in increased aerial contamination. Maximum aerial contamination occurred at indoor temperatures of between 65 and 75 degrees Fahrenheit and outdoor temperatures of approximately 50 degrees Fahrenheit. No relationship could be demonstrated between aerial contamination and relative humidity.

Specific environmental factors influenced by the duration of the

operative procedure which were studied included masking of personnel, splash basins, operating table mats and sheets. Masks were sampled by culturing 0.05 square inch cloth discs sewn on the inside and outside of the masks. 150 masks had an average of 11,173 organisms per 0.05 square inch on the inside and 454 on the outside. Respiratory organisms were shown to pass through the masks on only 3 occasions. Control masks sampled before use had an average of 88 organisms per 0.05 square inch on the inside and 733 organisms per 0.05 square inch on the outside. Splash basins were sampled at half hour intervals during the operative procedure. 103 of 122 basins remained sterile or the number of organisms present was too few to count. Fourteen of the remaining 19 basins had an average of 146 organisms per ml. The other five were heavily contaminated with an average of over 28,000 organisms per ml. Operating table mats and sheets were sampled by swabbing a one square inch area before and/or after the operative procedure. Mats had an average of 125 organisms per square inch before the operative procedure and 60 organisms per square inch after. Sheets had an average of 118 organisms per square inch before the operative procedure and 134 organisms per square inch after.

Factors affecting cleaning which were studied included scrub water for floors, mops, and floors. Sixteen samples of scrub water had an average of 74,237,500 organisms per ml. The majority of the organisms were of the coliform group. A progressive decrease in the degree of contamination during the day was shown. Twenty-one samples of one inch strands of mop fibre had an average of 6,528,560,000 organisms, the majority of which were coliforms. These counts also showed a progressive decrease in contamination during the day. Thirty-eight cultures of

operating theatre floors had an average of 346 organisms per square inch of floor surface. Twenty-eight cultures of operating theatre floors obtained after mopping had too many organisms to count. Three cultures obtained half an hour after the mopping, after the floors had dried, showed a much lower degree of contamination.

Factors concerned with anaesthesia which were studied included anaesthesia masks and endotracheal tubes. Masks sampled before use, after use and after storage showed averages of 910, 37,600 and 1909 micro-organisms per mask respectively. Pathogenic micro-organisms including _M. pyogenes var. aureus_ and _Pseudomonas pyocyaneus_ were among those which grew on the culture plates from anaesthesia masks. After a decontamination procedure was adopted average counts were 1489, 3000, 183 and 936 organisms per mask sampled before use, after use, after decontamination and from storage cabinets. Preliminary experiments showed that contamination could be transferred to the connecting tubes of the gas machine. Preliminary cultures of endotracheal tubes showed that growth could be demonstrated on only 1 out of 12 samples before incubation in broth, but growth was obtained from 9 of the cultures after the tubes were incubated. Cultures of 36 endotracheal tubes sampled after storage had an average of 183 organisms per tube. Organisms which grew in the cultured tubes include _Pseudomonas pyocyaneus_, _M. pyogenes var. aureus_ and coliforms. After the decontamination and storage procedures were altered, 19 cultures of endotracheal tubes had an average of 100 micro-organisms per tube.

Incidental factors which were studied were stretcher blankets, various depots or "fomites" which might harbour micro-organisms, surgeons' hands, patients' skin and the normal respiratory tract flora of operating theatre personnel. Cultures of 72 blankets obtained by a

sweep-plate technique had an average of 132 micro-organisms per square foot of blanket. The time of sampling had no apparent effect on the degree of contamination. M. pyogenes var. aureus was present in 51 of the cultures. Pseudomonas pyocyaneus grew on one of the culture plates. 192 cultures of depots from 15 different locations in operating theatres had an average of 723 micro-organisms per square inch. Walls, floors and the base of the operating tables were most heavily contaminated with averages of over 1000 organisms per square inch. M. pyogenes var. aureus grew on 8% of the culture plates. Control depots sampled in the laboratory had an average of 943 micro-organisms per square inch. Floors, radiators, sinks and shoes were most heavily contaminated. M. pyogenes var. aureus grew on 19% of the culture plates. Preliminary cultures of surgeons' hands before scrubbing, after scrubbing and after the operative procedure had average counts of 3023, 125 and 103 micro-organisms per swab. Cultures of 196 surgeons' hands had an average of 974 micro-organisms per swab after the preoperative scrub. Three of 7 cultures obtained after a second scrub had too many organisms to count, while the remaining 4 had an average of 225 micro-organisms per swab. Cultures of 122 surgeons' hands obtained after the operative procedure had an average of 636 micro-organisms per swab. The results showed that preoperative scrubs resulted in a 95% reduction in the flora of surgeons' hands. Preliminary cultures of patients' skin before preparation, after preparation and after the operative procedure had average counts of 1707, 947 and 280 micro-organisms per square inch. An average reduction in flora of 45% was shown. When an alcohol-iodine treatment was used a further reduction was shown after the operative procedure (to 56 micro-organisms per square inch). When Zephiran was used the number had increased after

the operative procedure (to 1400 micro-organisms per square inch). Cultures of noses and throats of nurses, surgeons, internes, anaesthetists, orderlies, secretaries and students showed an average carrier rate of 44% for _M. pyogenes var. aureus_. 33% of the personnel were carriers of coagulase positive _M. pyogenes var. aureus_. The influence of duration of service was best shown among internes and surgeons, whose carrier rates increased with their sojourn in hospital.

The results of the investigation were discussed in relation to existing techniques and measures which might be adopted to reduce the degree of contamination.

APPENDIXES

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APPENDIX A

Special Apparatus and Procedures

1. General Electric Electrostatic Bacterial Air-Sampler

The G.E. Electrostatic Bacterial Air-Sampler (Cat. No. 5140140 G 4, No. 3219491), also called the Duplex Electrostatic Air-Sampler, was used for air sampling. This apparatus operates on a 115 volt 60 cycle A.C. circuit. A vertical section of the apparatus is shown in Figure 1 D (page 153). The apparatus consists of a box containing a blower and two high voltage rectifiers which have a D.C. potential of 7000 volts. Two metal plates are situated at the top of the box. The right plate is connected to the positive terminal of one of the rectifiers and the left plate to the negative terminal of the other rectifier. Petri plates containing the suitable medium are placed on the metal plates. Air is drawn through $\frac{3}{4}$ inch holes in the clear plastic cover which is lined with metal cones. The air passes over the surface of the medium, below the charged metal plates and out through a hole in the back of the apparatus. The air flow is regulated at 0.5 cubic foot per minute. The total number of organisms is equal to the sum of the organisms collected by the positive and negative plates.

The apparatus is pictured in Figure 1 C (page 154) along with the Inter-Matic interval timer which was used to control the samples obtained at 5:00 - 6:00 a.m. Circular aluminum plates $2\frac{1}{2}$ inches in diameter with $\frac{3}{4}$ inch legs were taped over both openings of the sampler to prevent the deposition of organisms on the culture medium when the machine was not in use. One of these is shown in place over the left opening in Figure 1 C (page 154).

The advantages of the G.E. Electrostatic Air-Sampler, suggested by the designers (Luckiesh, Holladay and Taylor, 1946) are as follows:

(1) High efficiency.

(2) Simplicity of operation.

(3) Use of standard Petri plates.

(4) Freedom from contamination which would require sterilization between sampling.

(5) Constant and known air rate.

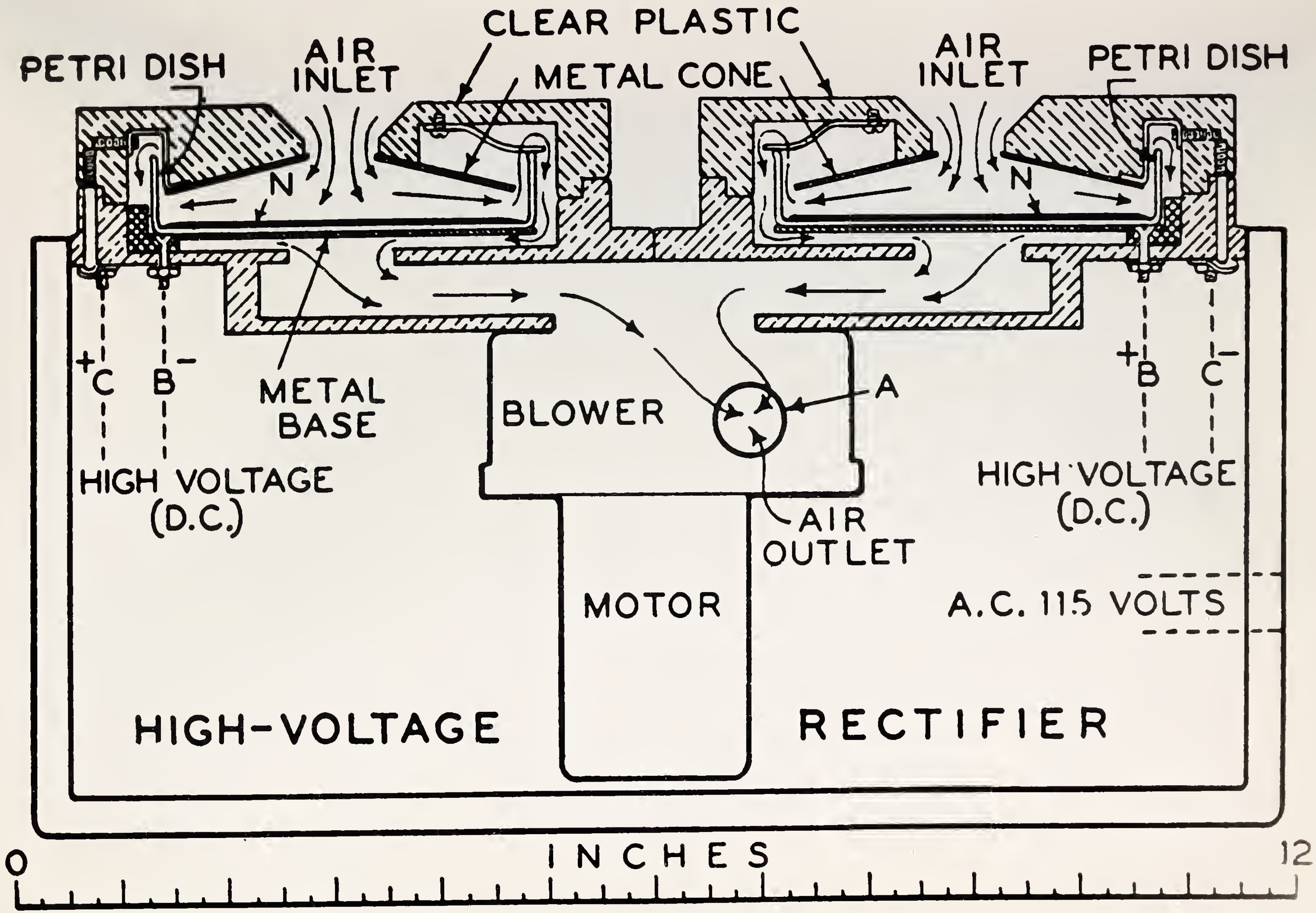
(6) Portable.

(7) Quiet operation.

(8) Freedom from hazards of high voltage electrostatic field (safety features of the sampler).

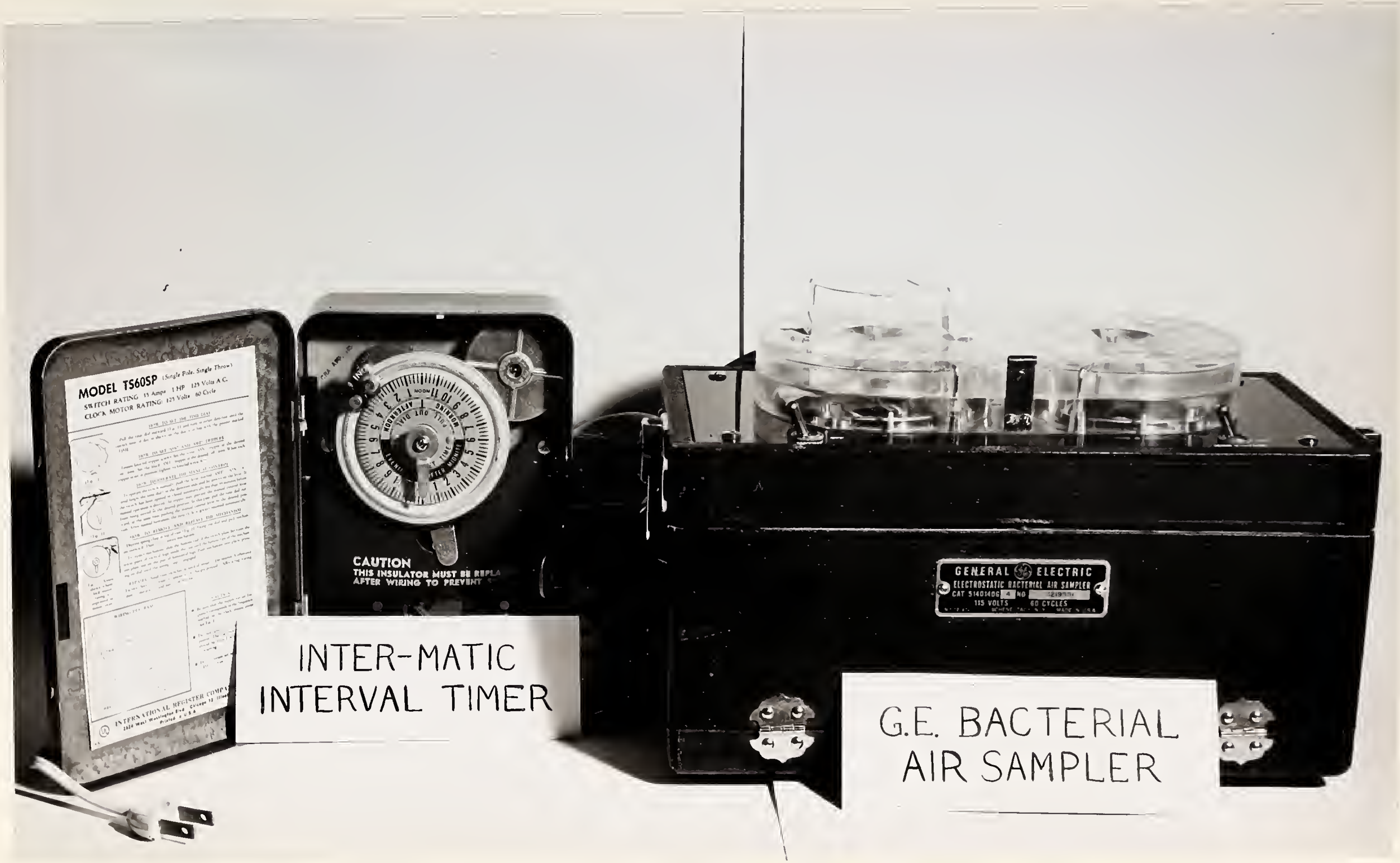
(9) Visual indicator - flashing lamp for electrostatic field.

(10) Operates on 110 to 120 volt 60 cycle alternating current.



A vertical section of the duplex electrostatic air-sampler

FIGURE 1C



General Electric Electrostatic Bacterial Air-Sampler and Inter-Matic Interval Timer

2. Correction of Relative Humidity Readings

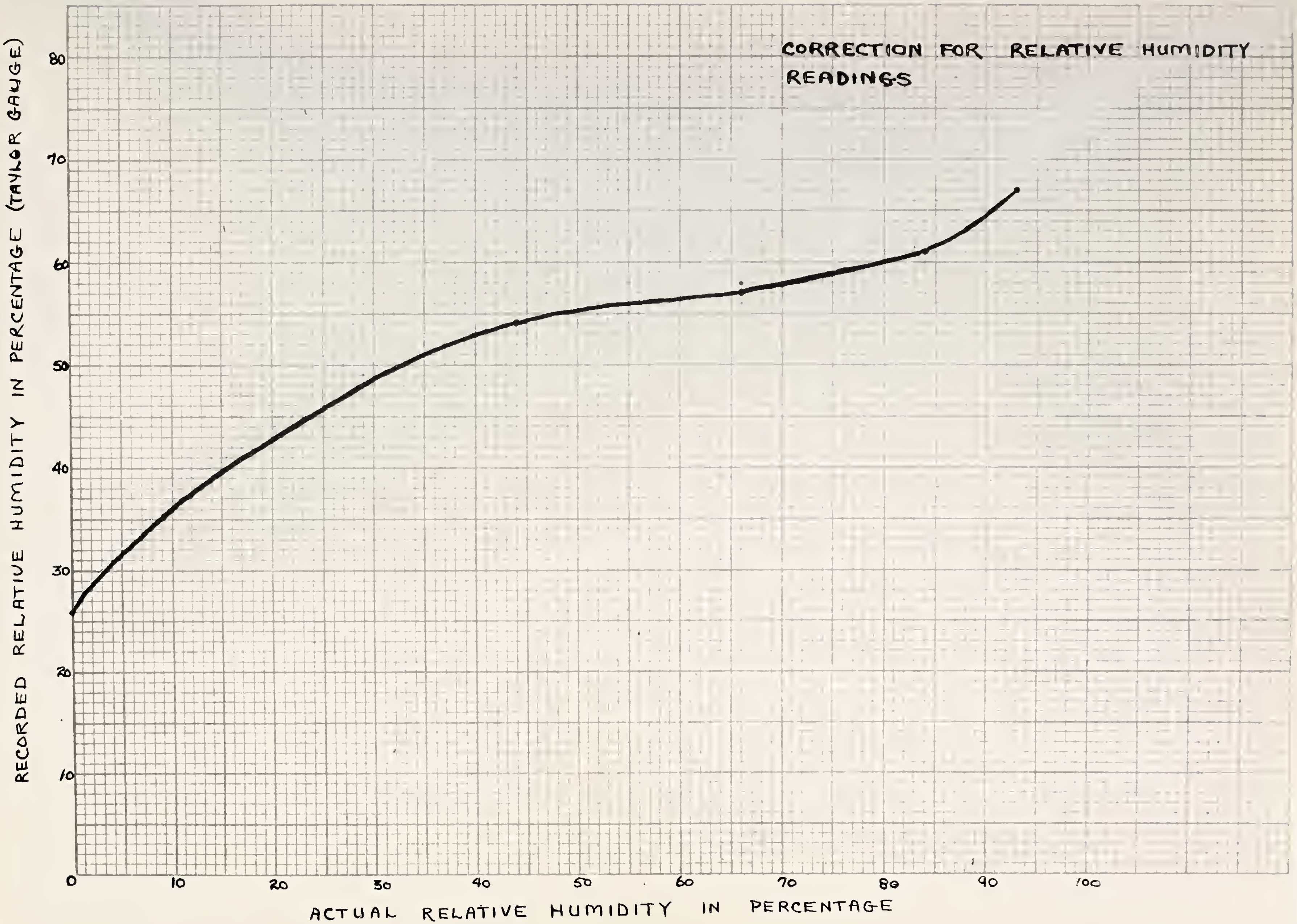
A Taylor "Humidiguide" humidity and temperature gauge was used to obtain relative humidity readings during air sampling. The readings were corrected by the procedure of Lidwell and Lowbury (1950). These workers used 9 inch sealed metal boxes with a capacity of 729 cubic inches containing anhydrous calcium chloride or 250 ml. saturated solutions of potassium carbonate, sodium nitrate, potassium bromide and sodium sulphate to obtain controlled relative humidities. Our procedure was modified by the use of a glass vacuum desiccator jar with a capacity of 629 cubic inches. Anhydrous phosphorous pentoxide was substituted for calcium chloride. The 250 ml. volumes of the saturated solution were placed in shallow glass bowls. The Taylor Humidiguide was placed in the desiccator jar along with the solutions. The jar was then sealed and set aside overnight to reach equilibrium before readings were taken. Actual and recorded relative humidities are given in Table 8 A (page 156). The results are presented graphically in Figure 8 B (page 157). This graph was used to correct all relative humidities given in Table 8 C (page 43) and Figure 8 D (page 44).

TABLE 8 A

Correction of Relative Humidity Readings

Substance used	Recorded Relative Humidity (Taylor Gauge)	Actual Relative Humidity
Phosphorous pentoxide (anhydrous)	26%	0%
Potassium carbonate (saturated solution)	54%	44%
Sodium nitrite (saturated solution)	57%	66%
Potassium bromide (saturated solution)	61%	84%
Sodium sulphate (saturated solution)	67%	93%

FIGURE 8B


CORRECTION FOR RELATIVE HUMIDITY READINGS
RECORDED RELATIVE HUMIDITY IN PERCENTAGE (TAYLOR GAUGE)
ACTUAL RELATIVE HUMIDITY IN PERCENTAGE
0
10
20
30
40
50
60
70
80
90
100

APPENDIX B

Media

1. Letheen Broth

Formula:

Lecithin (Eastman - animal)	Lot No. 7863	0.7 gm.
Tween 80 (Atlas)		5.0 gm.
Beef extract (Baltimore)	Lot No. 3856	5.0 gm.
Peptone (Armour - BDH)	Lot No. 05034	10.0 gm.
Sodium chloride		5.0 gm.
Distilled water		1000 ml.

Procedure:

Dissolve 0.7 gm. lecithin and 5.0 gm. Tween 80 in 400 ml. hot distilled water and boil until clear. Add 600 ml. of distilled water containing 5.0 gm. beef extract, 10 gm. peptone and 5.0 gm. sodium chloride. Boil this mixture for 10 minutes. Adjust the pH to 7 $\pm$ 0.2, filter through coarse filter paper, tube and autoclave for 15 minutes at a pressure of 15 pounds per square inch.

The broth was tubed in volumes of 10, 9, 4 and 3 ml. for use in culturing endotracheal tubes, splash basins, hands and anaesthetizing masks. This medium was recommended by Stuart et al. (1953) for neutralizing quaternary ammonium compounds. Rahn and Van Eseltine (1947) and Weber and Black (1948) reported the use of lecithin for neutralizing quaternary ammonium compounds. Baker et al. (1941) also used lecithin for inactivating quaternary ammonium. They believed that the polar molecules of the phospholipid lecithin acted by undergoing surface orientation on the bacterial cells, thus preventing the quaternary

ammonium compounds from exerting their effects on the cell wall and cellular contents. They also showed that lecithin must be added before or with the quaternary ammonium compound to be effective.

2. Sodium Thiosulphate Broth

Formula:

Nutrient (peptone) broth	1000 ml.
Sodium thiosulphate 10%	25 ml.

Procedure:

Add 25 ml. of a 10% solution of sodium thiosulphate in distilled water to 1000 ml. nutrient broth. Adjust the pH of the medium to 7 ± 0.2, tube and autoclave for 15 minutes at a pressure of 15 pounds per square inch.

The broth was dispensed in 4 ml. volumes in screw-capped vials. This medium was used for culturing the operative site after preparation with 2% Tincture of Iodine. The nutrient (peptone) broth was obtained from the Provincial Laboratory of Public Health.

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